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Original Articles

ON THE REAGIN CONTENT OF BLISTER FLUID*

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IT HAS long been known that certain antibodies are to be found in the fluid content of blisters. This was shown as early as 1897 by Hofmann (quoted by Thomas[2]) who recommended this fluid for use in the then recently discovered (1896) agglutination tests for typhus, and its use in the Widal test is mentioned by Jordan in his textbook of bacteriology.[1] Later, Thomas repeated this work[2] and determined the specific nature of the reaction. Buschke[3] has studied agglutinin and precipitin reactions in blister fluid and has used it in the Wassermann reaction. This latter was successfully repeated by von Wikullil.[4]

Recently, research workers in dermatology have been interested in the passive transfer of cutaneous sensitiveness to the normal from the individual demonstrating an allergic skin condition. The antibodies associated with the sensitiveness can be obtained, in many instances, in blister fluid, which, when introduced intracutaneously, sensitizes the normal skin at the site of injection. Fellner[5] transferred sensitiveness to tuberculin by the use of the fluid from the papule of a Pirquet's reaction; Urbach[6] with Konigstein used fluid from blisters, naturally or artificially produced, to transfer skin sensitiveness to arsenic, barley dust and arnica; while Perutz[7, 8] used the same method to transfer sensitiveness to turpentine. He apparently employed the contact test. Riehl[9] utilized the blister in transferring neosalvarsan idiosyncrasy.

It is, of course, well known that in other forms of clinical hypersensitiveness, such as bronchial asthma and hay fever, skin sensitizing antibodies are regularly present in the circulating blood, even though the principal shock tissues are located elsewhere than in the skin. It

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would be expected, then, that these antibodies are to be found in other body fluids, particularly in the tissue fluids of the skin of the hypersensitive individual. In conformance with this idea, and in view of the well-known presence of certain antibodies in blister fluids, we have studied the ability of cantharides blister fluid, obtained from the hypersensitive individual, to transfer specific sensitiveness to the normal skin. In this paper we have studied: first, the comparative content of skin sensitizing antibodies, or reagins, in blister fluid and in serum; and second, the specificity of the passive sensitization resulting from active blister fluid when injected into normal skins.

EXPERIMENTAL TECHNIC

Fluid from skin blisters was obtained by the cantharides method in a manner similar to that described by Petersen and Willis[10] in their studies upon capillary permeability.* Upon the anterior aspect of each thigh was placed a square of commercial blister plaster 1.5 cm. × 1.5 cm. in area, and covered with a square of adhesive plaster 3 cm. × 3 cm. Two blister plaster squares of the described size were employed, rather than a single square of twice the dimensions, to lessen the possibility of spontaneous or accidental rupture of the resulting blister membranes. The individuals so tested were instructed to remove the blister plasters at the end of six hours, and to cover the treated skin areas (the blisters having not yet appeared) with a celluloid vaccination shield or a square of gauze held in place with adhesive. In both sensitive and non-sensitive individuals, as shown by Petersen and Levinson,[11] the time interval necessary for blister formation is variable, being often as long as fifteen hours and seldom less than six hours; the occasional individual does not react at all to the test.

Upon the following day, using sterile precautions, the fluid is withdrawn from the blister into a 1 c.c. tuberculin type syringe, a 26 gauge needle being employed to pierce the blister membranes. Using cantharides plasters of the size described above, about 1 c.c. of fluid can be obtained from two blisters. The blister areas are then covered with a sterile gauze dressing. At this time blood for the comparative serum study is obtained by venipuncture. The blister fluid, after sterilization by filtration through a Seitz filter, is ready for use.†

Serum I, Blister Fluid I.—These were obtained from a late hay fever patient quite sensitive upon intracutaneous test to ragweed, giving a

*As indicated by these authors, cantharides, being lipoid soluble, readily enters the skin, probably producing its effect both on the sensory nerve endings and directly on the capillary wall. Being a specific capillary irritant without effect, to any extent, upon the rest of the protoplasmic elements, it allows the fluid to pass through the capillary wall. Wiedenfels, however (quoted by Pulay[12]), considers that the fluid is drawn from the lymph spaces. In either case the fluid must have passed through an endothelial wall.

†Since the smallest of the available commercial filters is much too large to accommodate the blister fluid specimens which vary in volume between 0.5 c.c. and 1.0 c.c., a special Seitz type filter of 2 c.c. capacity was devised for this study by Lorenz Westvig, Chief Engineer of the New York Post-Graduate Hospital, to whom we are indebted for his cooperation and interest.

marked reaction to a dilution of 0.001 mg. N per c.c. The blister fluid was obtained twenty-four hours after the six-hour plasters had been applied, the blood serum being obtained at the same time. Both

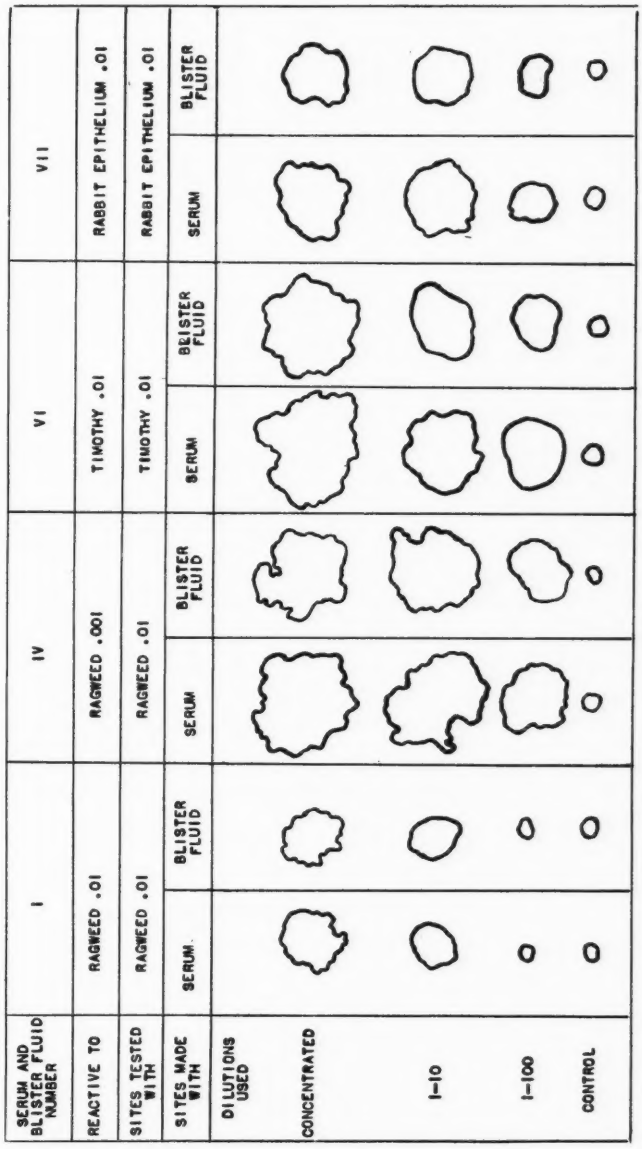

Chart 1.—Comparative results of the passive transfer to the normal human skin of serum and of blister fluid from the allergic individual.

fluids were Seitz filtered. Upon the following day sites were made in two normal individuals. This same procedure was followed with the remaining fluids.

Sera II, III, IV, V. Blister Fluids II, III, IV, V.—These were obtained from four late hay fever patients (II, III, IV, V) with a marked

TABLE I

COMPARATIVE RESULTS OF THE PASSIVE TRANSFER TO THE NORMAL HUMAN SKIN OF SERUM AND OF BLISTER FLUID FROM THE ALLERGIC INDIVIDUAL

SERUM AND BLISTER FLUID NUMBER	I		II		III		IV		V		VI		VII		VIII	
Reactive to	Ragweed 0.01		Ragweed 0.01		Ragweed 0.01		Ragweed 0.001		Ragweed 0.01		Timothy 0.01		Rabbit epithelium 0.01		Horse epithelium 0.01	
Sites tested with	Ragweed 0.01		Ragweed 0.01		Ragweed 0.01		Ragweed 0.01		Ragweed 0.01		Timothy 0.01		Rabbit epithelium 0.01		Horse epithelium 0.01	
Sites made with	Serum	Blister fluid	Serum	Blister fluid	Serum	Blister fluid	Serum	Blister fluid	Serum	Blister fluid	Serum	Blister fluid	Serum	Blister fluid	Serum	Blister fluid
Dilutions used																
Concentrated	Marked	Marked	Marked	Marked	Marked	Marked	Marked +	Marked +	Marked	Marked	Marked +	Marked +	Marked	Marked	Marked	Marked
1-10	Moderate	Moderate	Marked	Marked	Marked	Moderate	Marked	Marked	Moderate	Moderate	Marked	Marked	Marked	Moderate	Marked	Marked
1-100	Slight	Slight	Slight	Slight	Moderate	Slight	Marked	Marked	Moderate	Slight	Moderate	Slight	Marked	Moderate	Moderate	Moderate

Controls—All sites negative.

TABLE II

A COMPARISON OF THE SPECIFICITY OF THE SENSITIZING ANTIBODIES IN SERUM AND IN BLISTER FLUID

SERUM AND BLISTER FLUID NUMBER	REACTIVE TO	SITE NO.	TESTED WITH	READING 10 MINUTES LATER	ANTIGEN TESTED 25 MINUTES LATER	READING 10 MINUTES LATER
Serum I	Ragweed 0.01	1	Cottonseed 0.01	Negative	Ragweed 0.01	Marked
		2			Ragweed 0.01	Marked
Blister fluid I	Ragweed 0.01	1	Cottonseed 0.01	Negative	Ragweed 0.01	Marked
		2			Ragweed 0.01	Marked
Serum III	Ragweed 0.01	1	Cottonseed 0.01	Negative	Ragweed 0.01	Marked
		2			Ragweed 0.01	Marked
Blister fluid III	Ragweed 0.01	1	Cottonseed 0.01	Negative	Ragweed 0.01	Marked
		2			Ragweed 0.01	Marked
Serum IV	Ragweed 0.01	1	Cottonseed 0.001	Negative	Ragweed 0.01	Marked
		2			Ragweed 0.01	Marked
Blister fluid IV	Ragweed 0.01	1	Cottonseed 0.001	Negative	Ragweed 0.01	Marked
		2			Ragweed 0.01	Marked
Serum VI	Timothy 0.01	1	Cottonseed 0.01	Negative	Timothy 0.01	Marked
		2				
Blister fluid VI	Timothy 0.01	1	Cottonseed 0.01	Negative	Timothy 0.01	Marked
		2				

reaction upon intracutaneous test to ragweed 0.001 mg. N per c.c. with the exception of patient IV, a ragweed case, marked to ragweed 0.0001 mg. N per c.c.

Serum VI, Blister Fluid VI.—These were obtained from an early hay fever patient giving marked reactions to timothy 0.001 mg. N per c.c.

Serum VII, Blister Fluid VII.—These were obtained from a patient with bronchial asthma, aged thirty years, with marked positive reactions to house dust, orris and rabbit epithelium.

Serum VIII, Blister Fluid VIII.—These were obtained from a patient with bronchial asthma, aged twenty years, with marked positive reactions to house dust, horse epithelium and rabbit epithelium.

COMPARATIVE REAGIN CONTENT OF SERUM AND BLISTER FLUID

Dilutions of 1:10 and 1:100 in buffered salt solution were used of Serum I and Blister Fluid I reactive to ragweed, in the skin of two normal individuals and with the usual passive transfer procedure. One site each was injected with the concentrated fluids and their dilutions using 0.1 c.c. each. After twenty-four hours these sites and a control site in normal skin were tested with ragweed 0.01. In Chart 1 it will be seen that the reactions in the blister fluid sites are comparable to those obtained in the serum sites, though slightly less active. The lessened activity may denote a somewhat lowered reagin content in the blister fluid, or, what would seem more likely, a greater rate of dispersability of the fluid into the tissues after the sites are made. The same findings were obtained with Sera and Blister Fluids IV (reactive to ragweed), VI (reactive to timothy), and VII (reactive to rabbit epithelium). The remaining sera and blister fluids not depicted graphically in Chart 1 showed similar results (Table I). From these findings it would seem that the reagin content of cantharides blister fluid is quite comparable to that of the corresponding serum.

THE SPECIFICITY OF THE SENSITIZING ANTIBODIES IN BLISTER FLUID

Two sites each were made with the concentrated sera and blister fluids I, III, IV and VI, using 0.1 c.c. in each site, in normal individuals. At the end of twenty-four hours the first of each pair of sites was tested with an unrelated protein, cottonseed extract 0.01. No reaction developed by the end of thirty-five minutes in either serum or blister fluid site thus tested with a nonspecific excitant. The site and the hitherto unused control sites were then tested with the specific protein (Table II), a marked reaction developing in each case. The test tube neutralization method has not yet been attempted.[13]

CONCLUSIONS

The findings in this series of cases, admittedly small, would indicate that specific skin sensitizing antibodies regularly occur in the blister fluid as well as in the serum of many hypersensitive individuals.

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STUDIES ON POLLEN AND POLLEN EXTRACTS

XII. Enzyme Digestion of Pollen Allergens*

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THE digestion of pollen extracts with proteolytic enzymes has been carried out by a number of investigators, with what appear to be conflicting results.

Grove and Coca[1] digested pollen extracts with trypsin for twenty-four hours at room temperature, dialyzed in the refrigerator for three to six days, and found the product still active. Black[2] repeated this work and found the product quantitatively as active as the original by intradermal or intranasal test. On the other hand, Melli[3] was able to show that digestion with proteolytic enzymes under optimum conditions could cause a decrease in activity of pollen extracts. Bouillenne and Bouillenne[4] found that fifty days or longer were required for complete enzymic inactivation of ragweed pollen extracts, although a decrease in activity could be shown at the end of five days.

A critical consideration of the published results leads to the conclusion that the failure of the earlier workers to demonstrate inactivation of the allergic constituent in pollen extracts by proteolytic enzymes was due to lack of optimum conditions for the action of the enzyme and of a sufficient length of time to demonstrate a slow hydrolysis. It is hard to explain the complete disappearance of pollen nitrogen reported by Grove and Coca, a finding which has not been corroborated by any subsequent workers.

While it may be accepted that destruction of the pollen allergen is possible in the presence of proteolytic enzymes, the evidence does not appear conclusive that the loss of allergic activity is due solely to digestion of the proteins present. With our limited knowledge of the nature and ease of enzyme hydrolysis of pollen polysaccharides, and with the uncertainty regarding the action even of purified enzyme preparations, it appears at least not impossible that the enzyme preparations used might affect the hydrolysis of certain groups on the polysaccharide.

Our experiments were carried out in order to show whether enzyme digestion of pollen extracts might cause hydrolysis of the polysaccharide as well as of the protein. Presumably such hydrolysis should

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set free certain reducing groups on the sugars combined in the complex molecule. Acid hydrolysis readily causes the appearance of reducing sugars in an originally nonreducing (Fehling's test) solution of the polysaccharide.

EXPERIMENTAL

Commercial preparations of pepsin and of papain were used without further purification, as the purity of the enzymes used by some of the previous workers was unknown.

Pollen extracts were prepared by extracting with 100 c.c. distilled water for each 5 grams pollen used. Timothy and short ragweed pollens were used, as representative types of the two most important groups of hay fever producing pollens. Each of these extracts was used for the following experiments:

I. Pepsin digestion

Fifty c.c. pollen extract + 0.01 gm. pepsin (1:10,000) digested at 37° under toluene for seven days. Heated to boiling to destroy enzyme.

II. Papain digestion

Fifty c.c. pollen extract + 0.05 gm. papain digested at about 55° to 60° for six and one-half hours. Kept overnight in refrigerator and heated to boiling on following day to destroy enzyme.

Portions of each pollen extract were kept as controls. The P_H was not adjusted to the optimum for action of enzymes because it was feared that this might slightly influence the hydrolysis.

Samples of digests were used for testing on patients and for analyses for reducing sugars. Before use they were filtered to remove bac-

TABLE I

DERMAL REACTIONS OF HAY FEVER PATIENTS

PATIENT NO.		SHORT RAGWEED POLLEN				TIMOTHY POLLEN			
		ORIGINAL REACTION	CONTROL POLLEN EXT.	DIGEST		ORIGINAL REACTION	CONTROL POLLEN EXT.	DIGEST	
				I	II			I	II
(1)	I. P.	0	+	0	0	3+	4+	2+	0
(2)	F. B.	4+	4+	4+	4+	0	+	0	0
(3)	C. B.	4+	4+	2+	4+	0	0	0	0
(4)	N. B.	2+	2+	0	0	+	0	0	0
(5)	M. G.	0	0	0	0	4+	3+	+	4+
(6)	E. M.	3+	3+	0	0	0	0	0	0
(7)	H. B.	3+	+	0	0	0	0	0	0
(8)	M. T.	4+	4+	0	2+	0	0	0	0
(9)	G. E.	0	+	0	0	0	0	0	0
(10)	H. M.	3+	3+	+	3+	0	0	0	0
(11)	P. H.	−	0	0	0	−	2+	0	0
(12)	E. D.	4+	4+	2+	3+	2+	0	0	0
(13)	B. D.	4+	4+	+	0	2+	2+	0	0
(14)	M. H.	4+	2+	0	+	2+	0	0	0
(15)	C. H.	0	0	0	0	0	+	0	0
(16)	C. G.	4+	+	0	0	0	0	0	0

The first column under each pollen shows the reaction of the patient to routine testing in the clinic, and the second column the reaction to undigested control extract tested at the same time as the digested extracts.

teria. The samples tested on patients by the dermal test were mixed with an equal volume of glycerol. The results obtained are shown in Tables I and II.

TABLE II*

REDUCING SUGARS (AS DEXTROSE) MG. PER C.C.

	SHORT RAGWEED POLLEN	TIMOTHY POLLEN
Original extract	2.5	10.9
Digest I	2.75	10.3
Digest II	2.77	11.7

*The authors wish to express their appreciation to Mr. K. J. Bauer for these analyses.

DISCUSSION

From the tests on patients, it appears quite evident that the activity of the digested extracts is much less than that of the same extracts undigested. It appeared unnecessary to carry the digestion to a point at which the extract would fail to react upon the most sensitive skins. At the same time, the reducing sugars have not been increased, as might be expected if the polysaccharide were hydrolyzed. There remain the possibilities that the activity is due to a complex, the protein of which has been destroyed by proteolytic enzymes, leaving the polysaccharide intact, or that the protein alone is the active constituent. The former theory is made to appear improbable by the work of Stull, Cooke, and Chobot[5] and of Unger et al.[6] which indicate that the protein alone is capable of showing specific activity and that the pollen polysaccharides do not give the immune reactions, such as precipitin test, which are usually associated with such specific groups.

The conclusion then must be that the pollen allergen is a protein or protein-like substance, susceptible to hydrolysis by the ordinary proteolytic enzymes. The very slow rate of digestion may help to explain why such a high percentage of susceptible individuals becomes sensitized to pollens.

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ON THE "PROTEIN UNIT" STANDARDIZATION OF POLLEN EXTRACTS PROPOSED BY COOKE AND STULL*

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TWO questions concerning pollen atopens are now being investigated. The first pertains to the chemical nature of the atopen; the second, to its standardization in the usual extracts. The first question has not been experimentally examined in the present study, but occasional reference will be made to it in the course of this report.

Two methods of standardizing pollen extracts are now in general use; the weight per volume method and the total nitrogen content method. Under the former plan, the standardization is made either according to the Noon unit (the quantity of extract obtained from one-millionth of a gram of pollen), or in accordance with the dilution (1:100, 1:1000, 1:10,000) of the weight of pollen per volume of extracting fluid. The accuracy of the total nitrogen standard and the inapplicability of the weight per volume method have been clearly set forth in a recent paper by Coca.[1]

Another method of standardization of pollen extracts has been proposed by R. A. Cooke and A. Stull. These writers standardize their extracts according to the content of phosphotungstic acid (P-T-A) precipitable nitrogen; and they base this method upon their assumption that the P-T-A precipitable substance, which they call albumin, is the hay fever excitant of the pollen (pollen atopen).

In the paper mentioned above, Coca has confirmed the practical adequacy of the standardization by total nitrogen content and has presented evidence proving the method proposed by Cooke and Stull to be fallacious.

Coca tested the Cooke-Stull plan directly by comparing two timothy pollen extracts of different P-T-A precipitable nitrogen content as to their relative specific activity in timothy pollen hay fever subjects. He found that when these different freshly prepared extracts were standardized according to their total nitrogen content they exhibited approximately the same specific activity, notwithstanding the considerable difference between them with respect to their P-T-A precipitable nitrogen content.

The question arose whether these differences in P-T-A precipitable nitrogen content were of a magnitude which could be distinguished in comparative skin tests. This question was examined in the following manner. A single extract was diluted in two series, one series being

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made stronger than the other in the same ratio as the relative content of the P-T-A precipitable nitrogen in the two extracts mentioned above.

I received these dilutions not knowing which was the stronger series, and compared them by intracutaneous tests in hay fever subjects of different degrees of sensitivity. In each of these comparative tests the stronger solution elicited a definitely stronger reaction.

This direct test, which is obviously the final criterion of the practicability of the plan proposed by Cooke and Stull, had not been used by these authors. In their most recent paper they offer what seems to be an explanation for their having omitted this test, in the following passage:

"In many instances it was impossible to determine differences in strength of extracts by direct skin tests since frequently extracts 10 and even 100 times stronger by dilution, gave identical skin reactions when readings were made within thirty minutes. Only by testing a series of cases was it possible to determine the advantage of a protein nitrogen basis over a total nitrogen basis, and then only in the case of the orchard and timothy extracts, in which the variation in the percentage protein nitrogen approximated 30 per cent to 40 per cent."

In the present study this question has been taken up again with the use of the same method which was devised for the similar experiments that were reported by Dr. Coca. In principle this consists in carrying out the comparative tests on a number (about ten) of pollen sensitive subjects in dilutions that cause *less than maximal* reactions. This latter consideration is important because while it is easy to distinguish degrees of differences in slight and moderate positive reactions, it is sometimes difficult or impossible to detect degrees of differences in very marked positive reactions. It has been found disadvantageous also to compare the solutions in such weak dilutions that only very slight reactions are produced.

In the experiments reported by Coca and referred to above, decided differences in the same sense were obtainable with two solutions one of which was 50 per cent stronger than the other. The present study has shown that with the method described above it is possible to distinguish one dilution of a pollen extract from another dilution 25 per cent stronger but not from one 15 per cent stronger.

In the comparative tests carried out with two solutions of pollen extracts one 25 per cent stronger than the other, these were marked in such a way as not to indicate to the investigator which was the stronger. Sixteen comparative tests were made on the upper arms of eight sensitive subjects. In twelve instances, the reactions to the stronger solution were distinctly stronger than those to the weaker solution; in three instances, a difference in the reactions could not be distinguished, and in one the weaker solution produced the stronger reaction.

Comparative tests like those just described were carried out also with two solutions of an extract of Ascaris lumbricoides, one of which was 25 per cent stronger than the other (e. g., 0.01 and 0.0125 mg. N per c.c.). Sixteen comparative tests were made and in thirteen of these a distinctly stronger reaction was obtained with the stronger extract.

Finally, comparative tests were made with two solutions of histamine, one of which was 25 per cent stronger than the other (e. g., a 1:5000 and a 1:6250 solution). Twenty-two comparative tests were made, and in seventeen of these a distinctly stronger reaction was obtained with the stronger solution.

When two dilutions of the same ragweed pollen extract, one of them 15 per cent stronger than the other, were compared in 43 tests, the reactions were stronger in 17 instances with the stronger solution, equal in 11 instances, and stronger with the weaker dilution in 15 instances. From these results, although the tests were twice as numerous as in the previous series, it was impossible to judge which of the two dilutions was the stronger.

In view of the fact demonstrated above that two pollen extracts differing from each other in specific activity by only 25 per cent can be distinguished with properly conducted comparative skin tests, the plea of Cooke and Stull that their method of standardization cannot be subjected to the direct test employed by Coca seems not to be justified.

Cooke and Stull supported the basis of their method of standardization by reporting that a certain timothy pollen extract which had lost approximately one-third of its specific activity had lost also approximately one-third of its "protein nitrogen" content. In the paper by Cooke, Stull, and Tennant,[2] further evidence in the same sense is presented. A considerable loss of P-T-A precipitable nitrogen is stated to have occurred in eight different extracts during the period of one year.

In the present study, five ragweed pollen extracts of different ages (1925, 1927, 1928, 1930, 1931) have been compared as to activity and P-T-A precipitable nitrogen content with a fresh (1933) extract. The relative activity of these extracts was determined by skin tests in accordance with the method described above. The results of this study are shown in Table I.

According to Table I not even an approximate correlation was found between the relative specific activity of these extracts and their content of P-T-A precipitable substance.

It is still open to question, moreover, whether or not these extracts have suffered a perceptible loss of P-T-A precipitable substance. The content of this fraction in fresh extracts varies greatly. Thirty per cent is certainly not abnormal, and more pollen specimens must be

TABLE I

THE RELATIVE SPECIFIC ACTIVITY AND P-T-A PRECIPITABLE NITROGEN CONTENT OF RAGWEED POLLEN EXTRACTS OF DIFFERENT AGES COMPARED WITH A FRESH EXTRACT

	RESIDUE OF ACTIVITY PER CENT	P-T-A PREC. N PER CENT
1925	10	26.8
1927	30-40	23.5
1928	10	35.8
1930	20	46.0
1931	10	45.0
1933	100	40.0

examined before one can be certain whether or not 23.5 per cent, as found in the 1927 extract, can be assumed to indicate a "loss" of the P-T-A precipitable substance during the years since that extract was made. Stull, Cooke and Tennant mention one specimen (ragweed) which contained only 25 per cent. P-T-A precipitable nitrogen.

SUMMARY

1. Experiments are described which show that two dilutions of pollen extract and of an extract of Ascaris lumbricoides and of histamine differing in activity by 25 per cent can be distinguished from each other in appropriate comparative skin tests.

2. Two dilutions of the same pollen extract differing by 15 per cent in specific activity were indistinguishable in comparative skin tests.

3. The assertion of Cooke and Stull that the content of P-T-A precipitable substance in pollen extracts diminishes with time *pari passu* with the loss of specific activity could not be confirmed in a study of five pollen extracts from two to eight years old.

4. The present study confirms Coca's finding that the plan of standardizing extracts suggested by Cooke and Stull is fallacious.

The writer is indebted to Mr. E. L. Milford of the Lederle Laboratories, Pearl River, New York, for his kindness in carrying out the Kjeldahl determinations, upon the results of which the content of P-T-A precipitable substance was calculated for the pollen extracts referred to in Table I.

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A NEW DEFINITION OF THE NOON POLLEN UNIT

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IN 1911, Noon[1] announced the first application of the principle of specific desensitization in an allergic disease—hay fever. Realizing the necessity of some standard of dosage measurement, he adopted as the unit of such measurement the quantity of saline extract obtained from one-millionth of a gram of pollen. In laying down this unit, Noon was considering only timothy pollen, and he assumed that a certain weight of that pollen, wherever collected, always contains the same quantity of the hay fever excitant.

But even this latter assumption has been found by S. W. French[2] not always to be justified, and still larger discrepancies have been discovered between the weight of some pollens and their content of hay fever excitant.[2] For these reasons it would seem necessary to abandon the original Noon unit and to adopt another which would be uniformly applicable to all specimens of all the different pollens that cause hay fever. However, the preservation of this simple system of designating the dosage of pollen extracts will undoubtedly contribute to the wider use of the specific treatment of hay fever; and I have therefore thought it worth while to suggest a definition of the Noon unit which will accomplish this by correlating it with a chemical standard that has been shown to provide a reliable, indirect indication of the specific activity of the extracts of different specimens of widely different pollens: I mean the *total nitrogen* standard.

French[4] was the first to contribute experimental evidence of the reliability of the total nitrogen determination as a method of standardizing pollen extracts. He found that in successive extracts made from the same batch of pollen the total nitrogen content of the extracts paralleled their relative specific activity as determined by comparative skin and eye tests.

The results of more recent studies[5, 6] have testified to the superiority of the total nitrogen standardization over any other chemical method, and on the basis of these results one can formulate a chemical definition of the Noon unit which will make it a practically accurate standard.

The Noon unit can be defined as the *quantity of pollen extract which contains 0.00001 milligram of total nitrogen.* This definition actually applies to the original Noon unit of the usual timothy pollen extract. In other words, the extract from one-millionth of a gram of timothy

pollen usually contains very close to 0.00001 milligram of total N (a 1:100 extract contains about 0.1 mg. of total N per cubic centimeter).

For the convenience of those interested in the quantitative correlation of the Noon unit system, under the definition suggested above, with the two other current plans of determining dosage in the treatment of hay fever, the corresponding doses under the three plans are shown in Table I.

TABLE I

DOSAGE OF TIMOTHY POLLEN EXTRACT IN THE PROPHYLACTIC TREATMENT OF AN AVERAGE CASE (CLASS B) OF GRASS HAY FEVER (FIGURES IN THE THIRD COLUMN TAKEN FROM A. A. THOMEN[3])

DOSE	NOON UNITS	TOTAL NITROGEN MILLIGRAMS	WEIGHT PER VOLUME STANDARDIZATION		
			DILUTION	N PER C.C. MG.	EXTRACT C.C.
1	10	0.0001	1:10,000	0.001	0.1
2	20	0.0002	1:10,000	0.001	0.2
3	40	0.0004	1:10,000	0.001	0.4
4	70	0.0007	1:10,000	0.001	0.7
5	100	0.001	1: 1,000	0.01	0.1
6	200	0.002	1: 1,000	0.01	0.2
7	400	0.004	1: 1,000	0.01	0.4
8	600	0.006	1: 1,000	0.01	0.6
9	800	0.008	1: 1,000	0.01	0.8
10	1,000	0.010	1: 100	0.1	0.1
11	1,500	0.015	1: 100	0.1	0.15
12	2,000	0.020	1: 100	0.1	0.2
13	2,500	0.025	1: 100	0.1	0.25
14	3,000	0.030	1: 100	0.1	0.3
15	3,500	0.035	1: 100	0.1	0.35
16	4,000	0.040	1: 100	0.1	0.4
17	4,500	0.045	1: 100	0.1	0.45

It is seen that with the usual specimen of timothy pollen the figures are the same under the three plans though in different form. Thus, dose 17 is 4500 new Noon units, 0.045 milligrams of total nitrogen, or 0.45 c.c. of a 1:100 extract.

With most other pollen extracts, these figures would vary, and the only constant ones would be those in the second and third columns.

SUMMARY

It is suggested that the Noon unit be redefined, in accordance with its equivalent of specific activity in timothy pollen (which Noon used), as the quantity of pollen extract which contains 0.00001 milligram of total nitrogen.

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VISCERAL MANIFESTATIONS OF ANGIONEUROTIC EDEMA

Report of a Case with Recurrent Pulmonary Involvement*

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THE uncertain relationship between angioneurotic edema and allergy, and the fact that the magnitude of the rôle which allergy plays in the etiology and manifestations of disease is as yet only dimly apprehended, make it desirable to place on record all pertinent cases presenting new and unusual features in order that the deductions which must ultimately be drawn from the accumulated data may rest on a broad and secure foundation. The following case of angioneurotic edema, unfortunately incomplete, differs in several important respects from any that we have been able to find in the literature.

REPORT OF CASE

History.—The patient enjoyed good health from the time of his birth in 1917 until the spring of 1932, when without warning the dorsal surface of his left hand and forearm suddenly become swollen and itchy. These symptoms disappeared after a few days, but soon returned, and in the ensuing months there were a great many recurrences affecting the hands, feet, lips, and circumorbital regions. About January 1, 1933, he began to have headaches and shortness of breath which sometimes, but not invariably, occurred in conjunction with the subcutaneous swellings. The headaches were frontal in location, very severe, and not attended with nausea or vomiting. These attacks lasted several days, and recurred regularly at intervals of from seven to ten days. His suspicion that they were often accompanied by fever had never been confirmed. The most careful questioning failed to reveal any relationship between the attacks and specific foods or the multifarious plants and animals with which his work on a farm brought him into close contact. Obviously, the incidence was not seasonal. There was no history of hay fever, asthma, eczema, hives, migraine, or any other presumably allergic disturbance prior to the onset of the symptoms.

No information concerning the patient's immediate family could be obtained. Both his parents had died when he was a small child, and he had been separated from his brothers and sisters.

Physical Examination.—The patient was well developed and nourished. His mucous membranes were pale and his skin displayed a

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peculiar grayish pallor. The circumorbital areas and the dorsa of the hands and forearms appeared swollen, but pitting was not demonstrable. There were two red, circinate skin lesions, each 2 cm. in diameter, one on the right arm, the other on the abdomen. Many scratch marks, surrounded by secondary inflammatory reaction, on the back, arms, and wrists obscured whatever cutaneous lesions may have existed in these localities. The eyes, ears, nose, mouth, and pharynx were negative. The thyroid gland was not enlarged. Discrete, firm lymph nodes, varying in diameter from a few millimeters to two centimeters, were palpable in the posterior triangle of the neck, and in the axillae and inguinal regions.

The volume, density, and extensibility of the lungs were normal, and there were no râles. The heart was not enlarged, and there were no endocardial murmurs or abnormalities of the heart sounds. The arterial blood pressure measured 120/80 mm. Hg. Nothing abnormal was discovered in the abdomen or by rectal examination. There were no abnormal neurologic signs.

The patient presented many of the stigmata of hypopituitarism. His voice was still that of a child, there was little axillary or pubic hair, and he had experienced no sexual desire, erections, or nocturnal emissions. His skin was unusually dry and he perspired very little even in hot weather. The external genitals appeared normal.

Subsequent Course.—The patient was admitted to the University Hospital June 7, 1933. Three days later he developed severe headache and fever, and complained of shortness of breath. Examination revealed pronounced hyperpnea with sixty respirations to the minute. The localized edema previously observed had disappeared, and there were no new areas of swelling. The temperature was 104° F. and the heart rate 120 a minute. Scattered, moist, crepitant râles were heard in both lungs, but there were no demonstrable changes in pulmonary volume, density, or extensibility. Examination of the fundi showed nothing unusual. The hemoglobin was 60 per cent, the erythrocyte count 3,200,000, and the leucocyte count 22,500. Fifty-four per cent of the leucocytes were eosinophiles, 21 per cent polymorphonuclear neutrophiles, 16 per cent lymphocytes, and 6 per cent endothelial cells. The pressure of the subarachnoid fluid was 16 mm. Hg, which rose to 80 mm. Hg when the jugular veins were obstructed; the fluid itself was clear, colorless, and contained no more than the normal number of cells. Neither the removal of 20 c.c. of spinal fluid nor the intravenous injection of a hypertonic solution of glucose relieved the headache. Adrenalin administered intramuscularly reduced the respiratory rate on one occasion from 64 to 38 and made breathing easier, but did not alter the hyperpnea or the objective pulmonary signs.

On the morning of June 12 the patient still had headache and hyperpnea. His lungs were now full of crepitant râles, and the discovery here and there of small areas of somewhat questionable dullness to percussion, together with all the other clinical aspects of the case, strongly suggested early bronchopneumonia. Nevertheless, we were not prepared for the startling roentgenographic appearance throughout both lungs of a widespread infiltrative process such as is seen in fulminating tuberculous bronchopneumonia (Fig. 1). Forty-eight hours later the temperature, respirations, and heart rate had returned to normal, the headache had ceased, the râles had disappeared from the lungs, and the leucocyte count had fallen to 16,000. Roentgenograms made June 16 showed almost no trace of the extensive process which had been present four days earlier (Fig. 2), a

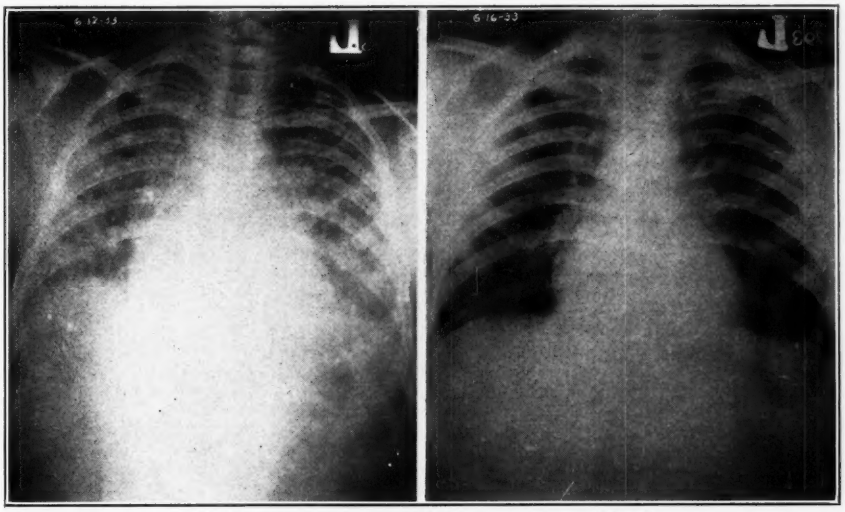

Fig. 1. Fig. 2.

Fig. 1.—June 12. Widespread process simulating tuberculous bronchopneumonia.
Fig. 2.—June 16. The evidences of intrapulmonic disease have disappeared almost entirely.

phenomenon no less startling than the original apparition. The patient declared that this episode was practically identical with those which had been occurring regularly during the preceding six months.

For ten days following the attack the patient remained free from subjective symptoms. A trace of albumin was found in the urine on two occasions, but not on a third. Chemical examination of the blood, including quantitative estimations of uric acid, urea nitrogen, creatinine, calcium, phosphorus, and plasma proteins, disclosed no abnormalities. The blood Wassermann and Kahn reactions were negative, as were agglutination tests for typhoid, paratyphoid, and undulant fever. The sella turcica appeared normal roentgenologically. The basal metabolic rate was normal. Unfortunately, the cutaneous lesions

on the back and arms made it impossible to perform skin sensitivity tests. The only remarkable things about the patient during this interim were his leucocytosis and eosinophilia; the number of leucocytes varied from 16,000 to 62,000, of which 54 to 84 per cent were mature eosinophilic polymorphonuclear cells. An axillary lymph node which was excised for histologic examination was found to be the seat of a simple chronic inflammatory process, presumably a result of the excoriative dermatitis.

On June 25 the patient's temperature rose suddenly to 103°F., but soon returned to normal. Roentgenograms of the lungs made the same day showed no abnormalities whatsoever (Fig. 3). On June 27 the fever returned, reaching 104°F., and was accompanied by severe headache and air hunger. Moist râles reappeared in the lungs, and cir-

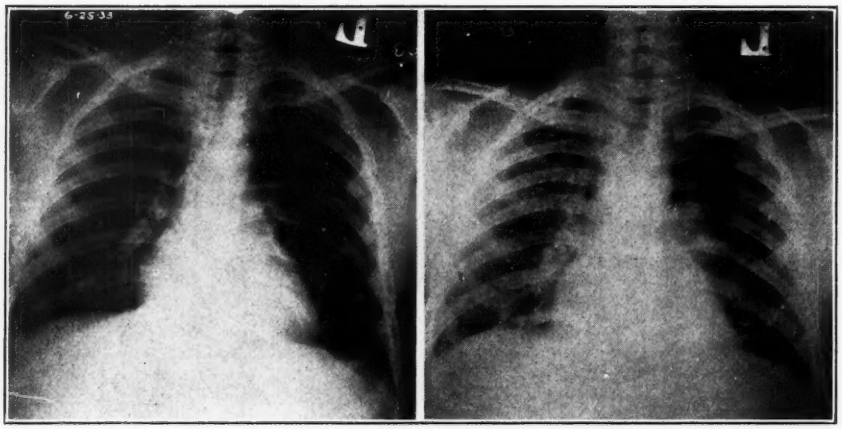

Fig. 3. Fig. 4.

Fig. 3.—June 25. The lungs continue clear.
Fig. 4.—June 27. The intrapulmonic lesions are returning.

cinate skin lesions similar to those present at the first examination broke out on the back and face. No localized edema appeared. Roentgenograms again revealed an extensive process simulating bronchopneumonia (Fig. 4).

The headache and respiratory symptoms, the latter practically unaffected by adrenalin, continued throughout June 28. Roentgenologically, the intrapulmonic process was found to have increased greatly in severity and extent since June 27 (Fig. 5). The respiratory rate fluctuated between 30 and 50 a minute. The leucocytes numbered 48,000, 80 per cent of which were eosinophiles. During the night the patient developed intense generalized pain and diffuse tenderness in the abdomen, together with some distention. Adrenalin afforded only partial, temporary relief from these symptoms. On the morning of June 29 the patient vomited repeatedly, and some of the vomitus was

bloody. Examination of the eyegrounds disclosed slight edema of the macular areas and evidence of detachment of the right retina. The headache, respiratory discomfort, and abdominal pain continued unabated until noon, when the patient died. It was impossible to obtain permission to make a post-mortem examination.

Summary.—History of many attacks of localized edema affecting the hands, forearms, feet, lips, and circumorbital regions, beginning in the spring of 1932. Onset seven or eight months later of attacks of severe frontal headache and shortness of breath (with fever?) which lasted several days and recurred periodically every seven to ten days. Presence, on admission, of questionable angioneurotic edema, erythematous lesions and scratch marks, and moderate enlargement of the cervical, axillary, and inguinal lymph nodes. No change of voice, no growth of axillary or pubic hair, no sexual desire, erections, or nocturnal emissions; dry skin, anhidrosis.

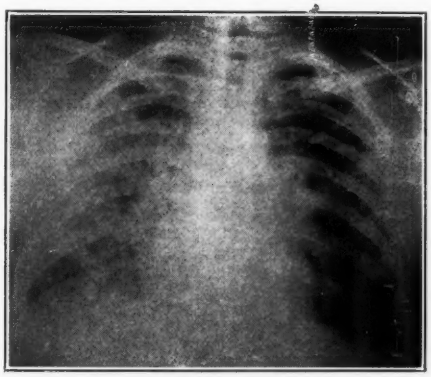

Fig. 5.—June 28. The process has increased greatly in severity and extent.

Subsequently, an acute febrile episode characterized by headache, tachycardia, shortness of breath, hyperpnea, tachypnea, pulmonary edema and the roentgenologic manifestations of bronchopneumonia, leucocytosis and eosinophilia.

A symptom free period of ten days, with persistent leucocytosis and eosinophilia. Histologic examination of an axillary lymph node disclosed a chronic inflammatory process.

A second febrile attack similar to the first, with, in addition, circinate skin lesions, severe abdominal pain and distention, vomiting, and hematemesis, ending fatally.

Diagnosis.—Cutaneous, cerebral, pulmonary, and gastroenteric manifestations of angioneurotic edema; hypopituitarism.

DISCUSSION

This case bears many striking resemblances to those of angioneurotic edema with visceral manifestations which have been described

by Osler, Fairbanks, Crowder, Christian, Crispin, and others. The nature of the attacks, with their sudden onset, rapid development of severe symptoms, and dramatic termination, together with their tendency to recur periodically, is characteristic of such cases. Osler, in discussing his "erythema group" of skin diseases, which included simple erythema, herpes iris, erythema nodosum, some of the purpuras, urticaria, and Quincke's edema, stated that all of them may be attended with visceral manifestations, and emphasized two points which are relevant: The first is that the visceral symptoms and the skin lesions may occur independently, and the second, that in any patient the skin lesions are frequently pleomorphic. Our patient did not have typical angioneurotic edema while he was under our observation, but his voluntary description of sudden transient swellings of the lips, eyes, and hands was unmistakable. Moreover, his family physician, who had actually observed these symptoms, had made a provisional diagnosis of "angioneurotic edema." The red, circinate lesions which were present when the patient came to the hospital and during the fatal attack were not unlike those described by Osler and by Herrmann.

Headache has been mentioned by practically all who have written on the subject as a frequent visceral manifestation of angioneurotic edema. As in our case, it is usually sudden in onset and termination, and very severe. It was assumed by Osler and other early writers that the headache and the symptoms of local encephalic lesions which occur in some cases are manifestations of intracranial edema. Evidence in support of such an assumption is to be found in the recent report by Bassoe of a case of angioneurotic edema in which the appearance of neurologic signs suggesting brain abscess led to surgical exploration and the discovery of nothing more than a localized area of edema and intense hyperemia. Lumbar puncture in Kennedy's case of urticaria with cerebral symptoms showed, as it did in our case, that the spinal fluid was under only slightly increased pressure and contained the normal number of cells.

Abdominal symptoms are among the most commonly observed of the visceral manifestations of Quincke's edema. They usually take the form of severe generalized pain and vomiting, and often include hematemesis, melena, distention, and rigidity. Surgical exploration in a considerable number of such cases, undertaken in the belief that the symptoms indicated the presence of a remediable lesion, has established the fact that localized edema of the intestine is the underlying pathologic process. The fact that a diagnosis of angioneurotic edema of the intestine does not *ipso facto* contraindicate surgical intervention is illustrated by a case of Sutherland's, cited by Osler, in which death occurred from intussusception of an edematous section of intestine.

The more unusual features of our case were the respiratory symptoms, fever, leucocytosis, and eosinophilia.

Angioneurotic edema of the respiratory tract is usually confined to the larynx and supralaryngeal structures. In only a few cases has involvement of the lower respiratory tract been suspected. Osler remarked the appearance and rapid disappearance of râles in one of his cases, and both Osler and Parker and Hazen mentioned pneumonia among the complications of Quincke's edema, but neither reported any necropsies, nor did they speculate on the nature of the pathologic process in the lungs. Vaughan and Hawke described a case of angioneurotic edema characterized by a great variety of visceral manifestations, including cough and pain in the chest, with roentgenologic changes suggesting pulmonary tuberculosis. The patient's respiratory symptoms were "promptly relieved by repeated injections of adrenalin, and a few days later roentgenograms showed entire disappearance of all suggestive lung findings." Feer's patient, after eating lobster, developed swelling in the throat and râles throughout both lungs without any indications of consolidation or asthma. The râles vanished within twenty-four hours, to be succeeded by typical angioneurotic edema of the face and arms. Feer's diagnosis was "Quincke's edema, primary in the lung." In this connection reference should be made to Löffler's cases of transient infiltration of the lungs. None of his patients had angioneurotic edema, and Löffler himself, in pondering the etiologic possibilities, did not mention angioneurotic edema or allergy. He inclined rather to the idea that the pulmonary signs indicated either fleeting pneumonia or incipient tuberculous infiltration. However, in view of the eosinophilia which was present in four of his five cases, and the increasing evidence that the infiltrative stage of pulmonary tuberculosis represents an allergic phenomenon,* it may be that these cases are at least partially comparable to ours.

Necropsies in fatal cases of angioneurotic edema are few, and information concerning the state of the lungs is meager. In Wason's patient, an infant who was asphyxiated by edema of the pharynx and larynx, the lungs "were voluminous and purple. . . . and pressure caused pink froth to exude into the trachea; microscopically there was edema involving the alveoli moderately, and the subpleural, peribronchial, and interlobar connective tissue markedly." In our patient, although we lack the proof that might have been obtained *post mortem*, it is practically certain that the respiratory symptoms and pulmonary manifestations were produced by Quincke's edema, for it is difficult to conceive of any other pathologic process which could have come and gone so quickly.

*See Spiro, P., and Becker, R.: Zur Pathogenese des Frühinfiltrats, Röntgenpraxis 2: 258, 1930.

With respect to fever, several cases comparable to ours have been reported, although as a rule the temperature does not rise very much above normal. In some of Osler's cases temperatures of 101°F. or 102°F. were observed, and in one instance 105° F. was recorded. Christian's patient, whose symptoms included purpuric skin lesions and pain in the joints and the abdomen, exhibited a temperature of 102.5° F. In Herrmann's case of erythema multiforme with visceral symptoms the temperature reached 104° F. Feer's patient, already referred to, was subject to urticaria and fever (104° F.) after sea bathing. Pesopoulos reported a case of Quincke's edema accompanied by high fever (105.8° F.), and quoted Cassirer to the effect that hyperpyrexia is observed occasionally in such cases.

In most of the earlier case reports, including Osler's, no leucocyte counts were given. Herrmann's case was the only one we could find in which hyperleucocytosis (42,000, 6 per cent of which were eosinophiles) was an outstanding feature. Crispin stated that one of his patients had eosinophilia, but he did not publish the differential count. Vaughan and Hawke's patient, whose symptoms were a manifestation of sensitivity to food, had an eosinophilia of 4 per cent. In Assmann's case of Quincke's edema and Raynaud's disease the eosinophiles increased to 10 or 15 per cent when edema appeared. In Löffler's Case 3 the eosinophilia reached 22 per cent.

The eosinophilia in our case (50 to 80 per cent) was so much larger than that which usually accompanies angioneurotic edema that one might hesitate to accept it as such unless other possible causes were clearly excluded. There is a remote possibility that the patient had scabies; some of his scratch marks looked not unlike the lesions characteristic of that disease. If so, it may have contributed to his eosinophilia. Trichinosis occurs sporadically in Iowa, and was not excluded beyond all question in this case because there was no autopsy or biopsy. However, the clinical course was unlike trichinosis, no muscular tenderness was present, and no history of eating uncooked pork could be obtained. Similarily, the possibility of intestinal parasitic infestation was not absolutely excluded, although several attempts to find ova in the feces were unsuccessful.

In the differential diagnosis of our case one more possibility remains to be discussed. We refer to the cases classified as eosinophilic leucemia or Hodgkin's disease with eosinophilic hyperleucocytosis, such as those reported by Bass, Hay and Evans, and Stewart, which are distinguished, *inter alia*, by leucocyte counts ranging from 26,000 to 200,000, with mature eosinophilic polymorphonuclear cells making up as much as 90 per cent of the total. Post-mortem examination in these cases has revealed either complete destruction of the architecture of the lymph nodes by eosinophilic infiltration, or the histologic changes characteristic of malignant lymphoma. In spite of the want of a

necropsy in our case, we feel reasonably sure that it did not belong in this group, not only because the clinical aspects were dissimilar, but also because histologic study of the excised axillary lymph node showed that the pathologic changes were inflammatory, that comparatively few eosinophiles were present, and that the structure of the node was intact.

The cause of the patient's unexpected and sudden death must remain a matter of conjecture. He was obviously not asphyxiated by edema about the glottis, nor did he pass into fatal uremia, as did several of Osler's patients. Inasmuch as his abdominal symptoms overshadowed all others toward the end, it seems reasonable to assume that some intra-abdominal crisis was chiefly instrumental in precipitating the fatal issue.

SUMMARY

Some of the visceral manifestations of angioneurotic edema are illustrated by the report of a case in which recurrent pulmonary involvement, high fever, and a predominantly eosinophilic hyperleucocytosis were the outstanding features.

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OBSERVATIONS ON THE CYTOLOGY OF THE SECRETIONS IN ALLERGY OF THE NOSE AND PARANASAL SINUSES*

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IT IS a generally known fact, that, in the various shock organs or tissues in which the lesion of allergy occurs, the pathologic picture is characterized by edema and eosinophilic infiltration. In the nasal, paranasal sinus, bronchial, conjunctival, intestinal and genitourinary tissues the eosinophiles migrate through the epithelium and may be found in the secretions. The demonstration of these cells in the secretions, therefore, may be utilized as a means of corroborating the diagnosis of allergy.

The earliest recorded description of eosinophiles was made by Warthin Jones[1] in 1846. A few years after the demonstration of Charcot-Leyden crystals in bronchial secretions in asthma by Leyden in 1872, Ehrlich[2] in 1879 described and named these leucocytic cells with acidophile granules, eosinophiles. In 1887, Bizzozero[3] described Charcot-Leyden crystals in the nasal secretions and called attention to the possible relationship of these crystals to the granules of eosinophile cells. This observation was confirmed by several authors, the most outstanding of whom were, Gollasch[4] (1889), Schmidt[5] (1891), Lewy[6] (1891), and Neusser[7] (1892). They emphasized the finding of eosinophilic infiltration and edema in the tissues and the demonstration of eosinophiles in the nasal secretions in cases of bronchial asthma, nasal polypi and spasmodic coryza. In 1895, Seiffert and Kahn[8] illustrated by means of color plates, the edema and eosinophilic infiltration in edematous polypi and in the nasal and paranasal sinus tissues in so-called acute and chronic catarrhal rhinitis and sinusitis. Since the primary observations on the demonstration of eosinophiles in the bronchial secretions, a great deal of significance has been attributed to these findings in asthma. Following the period of these earliest observations up until only a few years ago, however, these important and significant pathologic findings in the nose and paranasal sinuses either were not recorded or received no more than passing mention. Even in cases of frank nasal allergy described by the terms of nasal hydrorrhea, nasal neurosis, paroxysmal or spasmodic coryza, vasomotor or hyperesthetic rhinitis, the significance of edema and eosinophilic infiltration of the tissues and the presence of eosinophiles in the nasal secretions was not given consideration. During the twenty-five-year

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period from 1895 to 1920 there was much controversy and there were many divergent opinions expressed regarding the origin, pathology, and significance of nasal polypi. The most popular opinion and belief regarding nasal polypi was that infection was the chief etiologic factor.

The classification of the inflammatory diseases of the paranasal sinuses which has been recognized since the beginning of rhinology as a speciality, divides the various affections into three main classes: (1) the suppurative; (2) the nonsuppurative, or hyperplastic; and (3) the combined suppurative-hyperplastic diseases. This classification, however, does not recognize or distinguish the hyperplastic or polypoid diseases of allergic origin, but it is apparent that they have been included in the nonsuppurative and combined suppurative-hyperplastic groups. During the past few years the reports of Finck,[9] Mullin and Ball,[10] Hansel,[11] Coates and Ersner,[12] Weills,[13] Shambaugh,[14] Rackemann and Tobey[15] and others on the pathologic significance of edema and eosinophilic infiltration of the nasal and paranasal sinus tissues have shown that allergy plays an important and integral part in the diagnosis and treatment of the diseases of the nose and paranasal sinuses. The reports of Eyermann,[16] Leicher,[17] Hansel, Kahn and Stout,[18] and Johnson and Goldstein[19] on the significance of eosinophiles in the nasal and sinus secretions have emphasized the importance of considering the cytology as an aid in the diagnosis of allergy.

The revival and recognition of these pathologic findings have thrown renewed light upon the diagnosis and treatment of paranasal sinus disease and have emphasized the necessity of a reclassification of the inflammatory diseases of the paranasal sinuses. It is now apparent that practically all the nonsuppurative hyperplastic or polypoid diseases and many of the combined suppurative-hyperplastic diseases are of allergic origin. We must now consider the so-called inflammatory diseases of the paranasal sinuses as being (1) of infectious or suppurative origin; (2) of allergic origin; or (3) of allergic origin with superimposed infection.

By the comprehensive correlation of the clinical picture with the histopathologic examination of the tissues there should be no difficulty in establishing a definite diagnosis of paranasal sinus disease. It is not always practical, however, to perform a biopsy of the tissues, but the cytology of the nasal and sinus secretions may be considered an approximate index of the pathologic process occurring in the tissues. The cytologic findings, therefore, give important diagnostic information.

Whenever a patient is referred to the otolaryngologist by the allergist or the internist, the otolaryngologist should be capable of determining the presence or absence of the manifestations of allergy and he should be able to determine whether infection complicates an existing nasal allergy. Cases of headache and gastrointestinal disease of allergic origin often have mild or atypical nasal manifestations and these should be

recognized by the otolaryngologist. The rhinologist bases his diagnosis chiefly upon the clinical history, the nasal changes, the roentgenography of the sinuses, the bacteriology, the cytology of the secretions and the histopathology of the nasal and paranasal sinus tissues.

The following considerations on the cytology of the nasal and sinus secretions in allergy are based upon the observation of about 5,000 specimens of secretion from about 1,000 cases of nasal allergy in adults and children, associated in many instances with other manifestations of allergy, especially bronchial asthma. These observations were made over a period of about ten years, and many of the patients were repeatedly observed for several years during this time. The cytologic findings were correlated with the clinical histories, the skin tests, the x-rays of the sinuses, the bacteriology and other laboratory data, in order to make practical interpretations of the cellular reactions in the secretions. It is often quite impossible to draw conclusions from the examination of a single smear of secretion; therefore, it may be necessary to make an examination of several smears, especially if the specimen does not contain sufficient material or if acute or chronic infection complicates the picture.

In the collection of secretion from the nose for smear examination several methods may be employed. Specimens should be taken separately from each side of the nose. Secretion is most easily and readily collected by having the patient blow the nose on a waxed paper or cellophane handkerchief. This gives a specimen which represents only nasal secretion or a collection of both nasal and sinus secretion. If no secretion is available from blowing the nose it may be necessary to remove it by swabbing with a cotton applicator. Secretion may also be collected in a specimen bottle at the time of an acute exacerbation. The insertion of a saline tampon into the nose may stimulate the flow of secretion so that sufficient material may be obtained. Secretion from the individual sinuses may be obtained by aspiration or by puncture and washing. Gross masses of secretion may be used for smears or the returned fluid may be centrifuged if no gross mass of material is available.

In the collection of secretion for examination it must be taken into consideration that great variations may occur as to the quantity and quality of the material available. Patients usually report for examination during or immediately after an exacerbation of symptoms. During the periods of reactions the secretion may be thin and watery with very little if any mucus present. Thin watery material may show eosinophiles only after many examinations. Often they are found clumped in small masses or clusters. (Plate 1, *A*.) If mucoid material is obtained, an abundance of eosinophiles may be found. The histopathologic examination of the mucous membrane shows the maximum infiltration of the eosinophiles just beneath the basement membrane. From here they

migrate through the basement membrane and the epithelium to the surface where they are uniformly distributed. They are then collected in the mucous secretion which is rolled or propelled toward the nasopharynx by ciliary action. In this way the cells are collected in great numbers in the mucus. Watery secretion usually flows very freely and does not collect the cells as does the mucus. If the mucoid secretion should be very profuse, fewer cells may be found. Sometimes collections of mucus show very few if any cells. If one should happen to collect some material which had been immediately secreted from a gland, the cellular content may be very scarce because the eosinophiles do not migrate through the glands to any appreciable degree. Immediately following a period of reaction and during the quiescent stage which may immediately follow, an abundance of thick mucus may be found collected in the nasal cavity just as it collects in the bronchi after an exacerbation of asthma. This secretion usually contains an abundance of eosinophiles. During quiescent periods, however, the nose may be dry with very little if any secretion available for satisfactory examination. With the cotton applicator sufficient material may be obtained. One may find small bead-like masses which represent clusters of desquamated epithelial cells. Eosinophiles may or may not be found in them.

When a patient reports for observation during or following a period of acute exacerbation of symptoms, the question frequently arises as to whether the reactions were the result of allergy or the result of an acute infection. Sometimes the history suggests the nature of the reactions and sometimes it is quite impossible to determine without the examination of the secretion for the cellular response. The color or consistency of the secretion is frequently not an index of the cytologic contents. Clear secretion may contain many eosinophiles or many neutrophiles or it may contain varying proportions of both types of cells. Yellowish secretion, usually contains a marked predominance of neutrophiles but may contain a marked predominance of eosinophiles.

The staining technic used for preparing the smears for examination is the same as that used for blood smears. Smears may be fixed by drying or may be fixed by the application of methyl alcohol. We have used Wright's, Giemsa's and methylene blue eosin stains, but have found that Giemsa stain is the most satisfactory. Eosinophile granules stain a brownish red color and the nucleus stains blue. Neutrophiles show a clear violet cytoplasm with a deep blue nucleus. Giemsa stain is made fresh at the time of use. One drop of stain is diluted with one c.c. of distilled water. It requires about 1.5 c.c. of stain for each slide, which should be fully flooded. The stain should be allowed to remain on the slide about twenty or thirty minutes when it should be poured off and the slide gently dipped in distilled water. A few drops of ethyl alcohol may be poured over the slide to clear off excess stain. If the slide is stood

on end on filter paper, it drains and dries within a few minutes and is then ready for microscopic examination. Thick, grossly visible areas on the slide may be examined first with the low power then with high power or oil immersion. In the examination of the stained smear the eosinophiles may be noted with their characteristic granules. If stained too long or heavily, they may appear quite dark. It is very important not to mistake neutrophiles for eosinophiles. Neutrophiles contained in dense masses of mucus may undergo false staining, probably because of degeneration. They may take the eosin stain in excess and appear quite like eosinophiles, but upon close examination it will be noted that true granules are not present. The cytoplasm sometimes stains very faintly but the nuclei usually stain well. The margins of thick collections usually stain well. Smears should be spread well over the slide to avoid thick collections. The eosinophiles may be noted to float on the surface of the mucus.

In the interpretation of the cytology of the secretions one must take into consideration the type and quantity of secretion in the specimen and to correlate the findings with the clinical history and nasal changes. The clinical type of allergy under consideration and the possible relationship of acute and chronic infection must always be evaluated. An appreciation of the significance of the cytology of secretion can be accomplished only by individual experience, and then only after the examination of many hundreds of specimens. If one has this work done by a laboratory or by a technician the clinician sacrifices the real value of the cytologic findings, for individual interpretation must be correlated with all the clinical facts.

In the inspection of a smear, one containing only eosinophiles for instance, it will be noted that the cells are irregularly distributed. Sometimes they are scattered, arranged in clumps or arranged in streaks. (Plate I, *A, B.*) When mixed with neutrophiles they may likewise be irregularly distributed. Only certain parts of the slide may show significant areas. It is not unusual, for instance, to find a slide completely covered with neutrophiles except in certain small areas where eosinophiles are clumped in masses.

SCHEME FOR RECORDING THE CYTOLOGY OF NASAL SECRETIONS

E +−	Few scattered eosinophiles
E +	Many eosinophiles—may be clumped
E ++ +++ ++++	Eosinophiles very numerous—large clumps
E +− N +− +	Few scattered eosinophiles and neutrophiles
E +− N ++ +++ ++++	Few eosinophiles and numerous neutrophiles
E + N +− +	Many eosinophiles and neutrophiles
E + N ++ +++ ++++	Many eosinophiles and numerous neutrophiles
E ++ +++ ++++ N +− +	Eosinophiles very numerous—many neutrophiles
E ++ +++ ++++ N ++ +++ ++++	Eosinophiles and neutrophiles very numerous

In recording the cytologic findings the question arises as to what percentage of eosinophiles constitutes a diagnostic number. In view of the irregularity of distribution, it is quite impossible to make an accurate evaluation in terms of percentages. One can learn only by individual experience how to evaluate the cytology. In making an accurate record of the cytologic findings on a history the best method is a colored photograph, but since this is not practical, the next best method is the use of the terminology employed in recording the results of skin tests. The plan was adopted, therefore, of recording the cytologic findings in terms of plus-minus, 1, 2, 3, and 4 plus eosinophiles and neutrophiles. Although many epithelial cells may be noted in specimens, we have attached no definite significance to them. In using this plan of recording the cytology, due consideration must be given to the type of specimen under examination. A scanty amount of secretion may show a small but significant number of eosinophiles, while a good specimen may show a four-plus number. Fig. 1 shows the method of recording the cytologic findings in terms of eosinophiles and neutrophiles. Plate I illustrates four representative smears.

The presence of large numbers of eosinophiles in the secretions during quiescent, apparently symptom-free periods indicates that mild reactions may occur in the mucous membrane without definite symptoms. Patients, therefore, with almost symptom-free seasonal and nonseasonal allergy and with symptom-free asthma may show eosinophiles in the nasal secretions. During quiescent periods, between exacerbations, a large percentage will show no typical changes in the nasal mucous membrane. The membrane may appear quite normal.

In secretion containing both eosinophiles and neutrophiles the latter represent superimposed acute or chronic infection. In evaluating the numbers of neutrophiles in the secretion one must take into consideration that the neutrophilic response is always greater than the eosinophilic response and that the number of neutrophiles usually outnumbers the eosinophiles about 10 to 1. A plus-minus or a one-plus number of neutrophiles represents about ten times as many eosinophiles. In a smear with four-plus neutrophiles the field is completely covered with these cells.

During the winter months, when infections are more prevalent and the nose is subjected to environmental influences, a few neutrophiles are found even in normal noses. The presence of certain anatomic changes, such as those which cause obstruction and excessive dryness, may be responsible for the presence of a few neutrophiles at any time of the year. Age is often a factor, neutrophiles being found more frequently in older patients. The marked obstruction present in the nose during periods of severe reactions, causes the retention of secretions and predisposes to their stagnation and secondary infection with re-

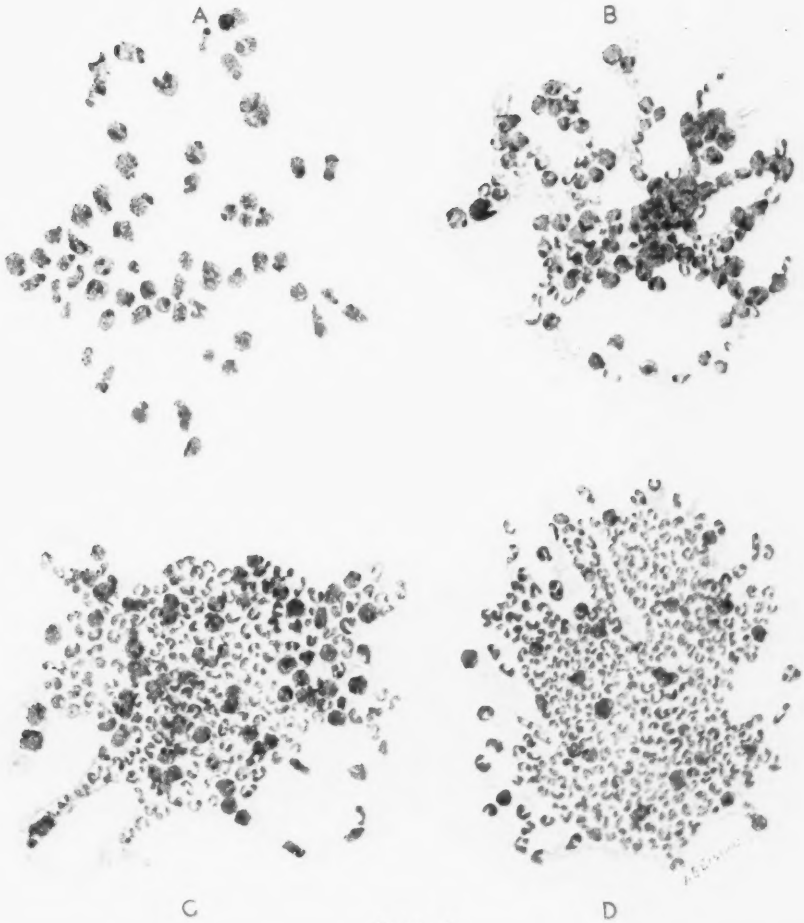

PLATE I

Nasal secretion. Giemsa stain, ×500. Four representative fields showing eosinophiles and neutrophiles. All diagnostic of allergy.

A. Eosinophiles arranged in a cluster. These clusters are frequently found in thin watery secretion but may assume this arrangement in association with neutrophiles.

B. Eosinophiles with a few scattered neutrophiles, enmeshed in streaks of mucus. This arrangement is most frequently observed.

C. Eosinophiles and neutrophiles, with a slight predominance of the latter. This picture is observed in the resolution stage of acute infection or in chronic infection; commonly noted in polyp cases.

D. Marked predominance of neutrophiles. Approximate diagnostic minimums of eosinophiles. Usually seen in complicating acute infection, but may be noted in chronic infection. This number scattered over several fields often observed in the resolution stage of acute infectious rhinitis in nonallergic patients, especially children.

sultant neutrophilic response. In cases of seasonal hay fever in which marked obstruction is present, there may be a number of neutrophiles present. In cases with hyperplasia of the membrane in which a great deal of the ciliated epithelium is replaced by the nonciliated types, the secretion tends to remain in the nose for abnormal periods of time and undergo secondary infection. The greatest degree of stagnation and secondary infection is found in cases of nasal polypi. In many cases of nasal polypi, however, the secretion may contain a marked predominence of eosinophiles. In these polyp cases the cytology of the secretions is an index of the underlying pathologic process and may be the deciding factor in determining the presence or absence of complicating acute or chronic infection. If acute infection is present, repeated smears should be observed over a period of several weeks before a definite decision should be made regarding the existence of the chronic type of infection. The persistence of a marked predominance of neutrophiles indicates chronic infection. In cases of acute infectious colds in nonallergic patients eosinophiles may be noted in the secretions mixed with the neutrophiles in the resolution stage of the infection. The number of eosinophiles may closely approach the number found in allergic patients during the acute stages of infection. Repeated follow-up smears must be made to make the distinction between the two types of cases. In the nonallergic patient the eosinophiles will disappear before the neutrophiles. In the case of allergy the number of eosinophiles will increase as the neutrophiles disappear until a pure eosinophilic response is again present provided chronic infection does not complicate the picture. An acute exacerbation of allergy may so closely simulate an acute cold that it is quite impossible in many instances to determine on the basis of the clinical history, which type of reaction has occurred.

The question frequently arises in cases of asthma, in both children and adults, as to whether an exacerbation of symptoms is caused or was caused by an infection or by a contact with some allergic substance. This may be readily determined by the examination of the secretion from the nose and treatment may be regulated accordingly.

Storm van Leeuwen and von Niekerk[22] have observed that there is no definite relationship between the percentage of eosinophiles in the blood and the clinical symptoms in allergy cases. In the secretions there is likewise no definite relationship to the blood eosinophilia. Marked fluctuations in the blood eosinophilia may occur when those in the secretions are fairly constant in number.

In cases with atypical symptoms and with no characteristic changes in the nose or in cases with thickening or polypoid changes in which there is a question of the existence of allergy, the cytology of the secretions must be relied upon to establish definitely the diagnosis. If acute infection is present with no eosinophiles the infectious process must be

allowed to subside before making a diagnosis. The repeated failure to find eosinophiles usually fits in with the absence of definite evidence of allergy.

DISCUSSION

In the consideration of the possible part played by bacteria in allergy of the nose and paranasal sinuses, particularly in those cases with hyperplasia and polypoid changes, the question has been recently brought up by Grove and Cooke[20] and by Kern and Schenck as to the possibility of bacterial allergy. Grove and Cooke believe that the hyperplasia and polyposis are caused by primary bacterial infection in an individual with an allergic constitution and that it represents a sensitization to infection. The finding of bacteria in the tissues in many of these cases and the observation of acute edema in the maxillary sinus following the subcutaneous injection of autogenous vaccines were the chief bases for their contention. Kern and Schenck state that "although it has been impossible to trace an exact parallel immunologically between atopy and so-called bacterial hypersensitiveness, upon closer study it would seem that the differences between the two states are rather matters of degree than kind." While more evidence is necessary, Kern and Schenck feel that it is but a step from this position to the assumption that bacterial allergy explains the mode of action of bacteria in the production of mucous polyps. Kern and Schenck believe that in the majority of cases the protracted action of allergens produces a chronically swollen mucosa (an allergic swelling). The subjection of this swollen mucosa to respiratory infections throughout the year, they state, prepares the way for bacterial hypersensitivity. The further explanation is offered that in the minority of cases there occurs first an infection with an infectious swelling of the mucosa and eventually if the patient is of allergic strain, there can supervene a true bacterial hypersensitiveness. This latter it is assumed will keep the mucosa in a swollen state, but the swelling is then no longer inflammatory (red, congested), but allergic (pale, edematous, hyperplastic). They finally conclude that the edematous mucous polyp is, therefore, the ultimate degree of allergic change in the mucosa.

It has been our experience that whenever marked hyperplasia and polyposis develop the membranes tend to become infected and invaded by bacteria. The neutrophilic response in the secretion in addition to the eosinophilic response is an indication of this infection. That bacteria may be present first in the mucous membrane and be the primary cause of the precipitation of an allergic condition is suggestive in some cases. We have repeatedly observed cases, however, in which, for instance, the manifestations of allergy in the nose were precipitated by an acute upper respiratory infection of acute sinusitis, yet these cases did not appear to be different from the other pure allergic types. Some

of these cases developed polyposis and some did not. We have not felt justified in assuming that a persistent bacterial allergy played a part in cases with a pure eosinophilic response in the secretions. Very few if any bacteria were noted in secretion with pure eosinophiles, but many were usually found in the presence of neutrophiles. Cases with neutrophilic response indicating chronic infection in which surgery was necessary to remove this infection responded on the basis of the removal of the ordinary inhalants and foods or on the basis of desensitization treatment (pollens, house dust, etc.). The results obtained on this basis of therapy in our experience are rather against the acceptance of a pure bacterial allergy. On the basis of our present knowledge of allergy, we believe, therefore, that a pure eosinophilic response in the secretions indicates an allergic process of an atopic nature and that this reaction is maintained by the ordinary allergens. That a pure eosinophilic response is caused by a bacterial allergy is difficult to prove. Clinical evidence does not substantiate this belief. In a recent contribution on the subject of infective asthma and its allergic nature, however, Cooke reported that eosinophilia may be regarded as a bacterial sensitization phenomenon equally as well as it is so regarded in serum disease and experimental anaphylaxis. When there is an acute or chronic infection, the neutrophilic response in the secretions is according to clinical evidence, an indication of the existence of infection. Certainly in acute infections in allergic patients who previously showed only eosinophiles in the secretions, the immediate neutrophilic response is proof of this infection. The persistence of neutrophiles in large numbers in addition to a certain number of eosinophiles has according to our experience substantiated the belief of the existence of chronic infection.

Repeated respiratory infections in cases of polyposis seem to play a part in increasing the edema, but the primary cause of polyposis appears to be allergic edema. In many cases with repeated infections occurring over a period of many years polyposis has never occurred. In our series of cases there is no definite relationship between the onset of the nasal manifestations of allergy with acute upper respiratory infections and the eventual development of polyposis. In a very significant percentage of cases of polyposis the nasal manifestations of allergy began primarily as hay fever or pollinosis. A detailed report on the relationship of bacteria and bacterial allergy is now in progress and will soon be published.

SUMMARY

1. The demonstration of eosinophiles in the secretions from the nose and paranasal sinuses is good presumptive evidence of the existence of allergy.

2. The presence of neutrophiles in the secretions in addition to eosinophiles is an indication of the existence of superimposed infection.

3. The eosinophilic-neutrophilic proportions are an index of the nature of the infection.

4. By repeated observations on the cytology of the secretions, acute and chronic infections can be differentiated.

5. A pure eosinophilic response as the result of bacterial hypersensitiveness is not definitely substantiated on the basis of clinical evidence.

6. Cases of hyperplasia and polyposis with a persistent absence of a diagnostic number of eosinophiles, as a rule, do not prove to be allergic.

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COMPARISON OF REAGINS TO SEPARATE SPECIES OF CADDIS FLY

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AS REPORTED in several articles, Parlato has shown that in the vicinity of Buffalo certain individuals show clinical hypersensitivity to emanations of the Caddis fly, that extracts of the whole fly will produce positive reactions on intradermal and ophthalmic tests in these individuals, and that reagins to the Caddis fly allergen can be demonstrated as present in their blood serum. Subsequently he reported finding reagins to moth and butterfly extracts in certain individuals. He compared these reagins with that to Caddis fly by means of the so-called exhaustion of passive transfer sites, and concluded that the reagins to butterfly and moth are identical and that the reagin to Caddis fly is similar to that of moth and butterfly.

There are several varieties of Caddis fly found around Buffalo. Two of them are more abundant than the others: a small brownish variety, *Hydropsyche chlorotica* Hagen *seu alternans* Walker, and a larger variety, *Macronema zebrata* Hagen. The former is the more plentiful. Disregarding the reported similarity of Caddis fly, moth and butterfly reagins, it seemed of interest to compare the extracts and the reagins thereto, of these two species, by means of exhaustion or desensitization of passive transfer sites.

Identification of the species *Macronema zebrata* Hagen was made independently by Mr. William Wild, entomologist of the Buffalo Society of Natural Science, by Dr. F. M. Carpenter, Biological Institute, Harvard University, and by Mr. J. H. Roberts of the University of Maryland. The *Hydropsyche* lot was examined by the same entomologists; it appeared to be made up chiefly of the species *Hydropsyche chlorotica* Hagen with a minority of a very closely related subspecies *Hydropsyche alternans* Walker *seu morosa* Hagen.

Morphologically, there is a considerable difference between the species *Macronema* and the species *Hydropsyche*. The above two subspecies of *Hydropsyche* closely resemble each other. Inasmuch as the point of the investigation was to discover the similarity or difference between the main species, allergenically, it is not of great importance that the *Hydropsyche* lot was a mixture of two closely related subspecies.

METHOD

In the summer of 1932, I was fortunate to take a quantity of these two common varieties separately in practically pure lots. The *Hydropsyche* lot contained about 5 per cent of the *Macronema*, which were

*From the Allergy Clinic, Buffalo General Hospital.

carefully removed by sorting after drying. The *Macronema* lot was uncontaminated. The flies were taken alive by net, killed immediately by the liquid and fumes of carbon tetrachloride, air dried while protected from dust, sorted to remove the rare extraneous insects (gnats, May flies, etc.), defatted with petroleum ether and extracted with Coca's solution containing 0.4 per cent phenol. The separate extracts were sterilized by Seitz filtration, and the total nitrogen content was determined. Dilutions in buffered saline solution containing 0.4 per cent phenol were made on the total nitrogen basis.

Blood serum, which had been obtained from a patient (G. M.) clinically hypersensitive to Caddis fly, and which had been shown to contain reagins to Caddis fly, was used for passive transfer.

In preliminary testing by direct intradermal method of the two extracts on a Caddis-fly-sensitive patient (W. E.) it was found that dilutions of equal total nitrogen content were not of equal activity. By comparing the activity of several dilutions of the two extracts, a dilution of each was chosen which gave equivalent reactions on W. E. (see Table I) and these were used for the experiment.*

TABLE I

INTRADERMAL TESTS ON PATIENT W. E. TO DISCOVER DILUTIONS OF EQUIVALENT ACTIVITY

MACRONEMA EXTRACT		HYDROPSYCHE EXTRACT	
TOTAL N PER C.C.	REACTION	TOTAL N PER C.C.	REACTION
0.001	++±	0.0005	++
0.0025	++	0.001	++
0.005	++	0.005	+++
0.01	+++	0.01	+++±
Macronema 0.005 = *Hydropsyche* 0.001			

Five sites on the upper arm of three individuals, proved to be nonreactors to Caddis fly extract, were sensitized by injecting 0.2 c.c. of serum. Sites 1 and 2 (see Tables II, III and IV) were injected at two- to four-day intervals with about 0.03 c.c. of the *Macronema* extract; Sites 3 and 4 were similarly injected with the *Hydropsyche* extract. Site 5 was injected with our routine Caddis fly extract (S. F. 32) which was prepared from a mixture of *Macronema* and *Hydropsyche*, the latter predominating. The reactions were read after fifteen to thirty minutes.

When Site 1 no longer reacted to the *Macronema* extract, an interval of two to three days was allowed to elapse to avoid a "refractory pe-

*It should be noted that although both extracts were made up initially on the basis of the same weight of flies per volume of extracting fluid (7 gm. per 100 c.c.), the undiluted *Hydropsyche* extract contained 1.60 mg. total N per c.c., and the *Macronema* extract 1.17 mg. per c.c. Also subsequent comparison of the two extracts on the patient G. M. by direct intradermal tests showed that the 0.001 *Hydropsyche* dilution produced a somewhat stronger positive than did the 0.005 *Macronema* dilution.

Patient G. M. had been unavailable for skin testing at the time of the experiment because of pregnancy and confinement.

TABLE II

REAGIN EXHAUSTION IN PASSIVELY SENSITIZED SITES

SERUM G. M.					RECEPTOR H. O.					
DATE	SITE 1		SITE 2		SITE 3		SITE 4		SITE 5	
1/28	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		Ragweed	
	0.001	+	0.001	+	0.001	++++	0.001	++++	0.01	0
	Control	0			Control	0			Control	0
2/ 2	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	+++	0.005	+++	0.001	++	0.001	+++	0.001	++++
2/ 4	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	++	0.005	++	0.001	+	0.001	+	0.001	+
2/ 6	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	++	0.005	++	0.001	±	0.001	±	0.001	+
2/ 8	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	+	0.005	+	0.001	0	0.001	0	0.001	0
2/11	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	0	0.005	0	0.001	0	0.001	0	0.001	0
2/13	*Hyd.*		Goat		*Mac.*		Goat		S.F. 32	
	0.001	++	0.01	0	0.005	0	0.01	0	0.001	0
	Control	0	Control	0					Control	0
2/18	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	0	0.005	0	0.001	0	0.001	0	0.001	0
	Control	0			Control	0			Control	0

TABLE III

REAGIN EXHAUSTION IN PASSIVELY SENSITIZED SITES

SERUM G. M.					RECEPTOR F. S.					
DATE	SITE 1		SITE 2		SITE 3		SITE 4		SITE 5	
1/28	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		Ragweed	
	0.001	+	0.001	+	0.001	++++	0.001	++++	0.01	0
	Control	0			Control	0			Control	0
2/ 2	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	+++	0.005	+++	0.001	++++	0.001	++++	0.001	++++
2/ 4	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	++	0.005	++	0.001	+	0.001	+	0.001	+
2/ 6	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	±	0.005	0	0.001	0	0.001	0	0.001	0
2/ 8	*Hyd.*		Goat		*Mac.*		Goat		S.F. 32	
	0.001	+++	0.01	0	0.005	0	0.01	0	0.001	0
			Control	0						
2/11	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	0	0.005	+	0.001	0	0.001	0	0.001	0
2/18	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	0	0.005	0	0.001	0	0.001	0	0.001	0
	Control	0			Control	0			Control	0

riod," and the site then injected with the *Hydropsyche* extract. Similarly, after Site 3 had become desensitized to *Hydropsyche*, it was injected with the *Macronema* extract. Site 5, after it had become desensitized to the routine extract S. F. 32, remained so to the end of the experiment.

Control injections were made on unsensitized skin with all the extracts at the beginning and end of the experiment.

TABLE IV

REAGIN EXHAUSTION IN PASSIVELY SENSITIZED SITES

SERUM G. M. DATE	SITE 1		SITE 2		RECEPTOR C. N. SITE 3		SITE 4		SITE 5	
1/28	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		Ragweed	
	0.001	++	0.001	++	0.001	++++ (M)	0.001	++++ (M)	0.01	±
	Control	0			Control	0			Control	0
2/ 2	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	++++	0.005	++++	0.001	++++ (M)	0.001	++++ (M)	0.001	++++ (M)
2/ 4	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	+++	0.005	+++	0.001	++	0.001	++	0.001	+
2/ 6	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	+++	0.005	+++	0.001	+	0.001	++	0.001	±
2/ 8	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	0	0.005	0	0.001	0	0.001	0	0.001	0
2/11	*Hyd.*		Goat		*Mac.*		Goat		S.F. 32	
	0.001	++++	0.01	0	0.005	0	0.01	0	0.001	0†
	Control	0	Control	0	Control	0			Control	0‡
2/13	*Mac.*		*Mac.*		*Hyd.*		*Hyd.*		S.F. 32	
	0.005	0	0.005	0	0.001	+	0.001	0	0.001	0
	Control	0			Control	±			Control	0

†Delayed + after six to eight hours.
‡Delayed + after six to eight hours at control injection on unsensitized skin.

The passively sensitized sites on all receptors regularly reacted to both *Macronema* and *Hydropsyche* extracts. It is therefore evident that the serum of G. M. contained reagins to both species, although the reagins might be identical.

A positive reaction was regularly produced on the desensitized *Macronema* site (Site 1) by the injection of *Hydropsyche* extract. However, after desensitization, the *Hydropsyche* site (Site 3) failed regularly to react to an injection of *Macronema* extract.

Sites 2, 4 and 5 became desensitized respectively to *Macronema*, *Hydropsyche* and the mixed extracts, and remained so. The behavior of these three sites tends to show that the reaction produced by the *Hydropsyche* extract on the desensitized *Macronema* site was of a specific type, not irritative.

As noted in Tables II, III and IV, all control injections on unsensitized skin were negative in all receptors at the beginning of the experiment, and in receptors H. O. and F. S. again at the end. In receptor C. N. the slight delayed reactions on Site 5 and control at next to the last injection, and the slight immediate reactions on Site 3 and control at the end, may possibly indicate the development of a slight, fleeting hypersensitiveness to the extracts in this individual.

COMMENT

The above mentioned results may be interpreted by assuming that in the two species of Caddis fly studied, there are at least two different allergens present, the species *Macronema* containing one and the species *Hydropsyche* containing both; and that the serum of G. M. contains reagins to both of them. On this assumption the reactions can be explained as follows: *Hydropsyche* contains allergens X and Y, *Macronema* contains allergen X. Site 3 in the tables, which contains reagins X and Y is completely desensitized by the *Hydropsyche* extract, which contains allergens X and Y. It therefore does not react when the *Macronema* extract, containing only allergen X, is injected. On the other hand, Site 1, also containing both reagins, has only the reagin X exhausted by the *Macronema* extract, leaving the reagin Y still reactive to the corresponding allergen Y present in the *Hydropsyche* extract.

A further study of the two extracts should be made, using serum from other Caddis-fly-sensitive individuals, to check up on these results.

CONCLUSIONS

1. Reagins to two species of Caddis fly, *Macronema zebrata* and *Hydropsyche chlorotica seu alternans*, were demonstrated in the blood serum of a patient hypersensitive to this family of insects.

2. It is probable that the allergens present in the two species are not identical.

3. An explanation of the difference can be made by assuming that *Hydropsyche chlorotica* has two allergens, while *Macronema zebrata* has only one, and this one is identical with one of the two found in the former species.

I wish to express my appreciation to Dr. Roger S. Hubbard for the chemical analysis and to Dr. Stuart Vaughan for sterility tests on the extracts, also to the receptors for volunteering for the passive transfer tests, all of the Buffalo General Hospital.

REFERENCE

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THE URINARY PROTEOSE IN ALLERGY*†

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THE reports of Oriel and his associate[1] regarding the finding of a "proteose" in the urine of allergic persons aroused considerable interest and the hope that this might prove to be another avenue of approach to the solution of some of the problems associated with allergy. They found in all urines a nitrogenous material which could be separated by ether and precipitated by alcohol, but this substance was found in much greater amounts in those suffering from allergic conditions if the urine were collected during active manifestations of allergy. It was stated, also, that the proteose from the urine of the allergic person produced skin reaction in the patient from whom it was obtained and, in some instances, in others sensitive to the same allergen. No such reactions were obtained with the proteose from normal individuals. Although this method did not identify the allergen, it apparently offered a means of isolating it in those persons in whom it had been impossible to determine it by other methods.

Since it was believed that they were dealing with a specific substance, it was logical to attempt desensitization with it, and these workers reported successful treatment of various conditions some of which are not generally recognized as allergic.

Barber and Oriel[2] described the materials found as biuret, Millon and xanthoproteic positive and giving the Molisch reaction indicating a carbohydrate moiety. It was not coagulated by heat and was precipitated by lead acetate, alcohol and saturated ammonium sulphate. Hydrolyzed with hydrochloric acid a brown precipitate was obtained which contained phosphorus and was believed to be nucleic acid. They concluded that the proteose contains the specific antigen combined with some product of tissue autolysis.

Oriel[3] found that the proteose sensitized the uterus of the guinea pig and could desensitize it. A heterologous proteose gave partial contraction. Serum from a patient suffering from both asthma and eczema injected into a guinea pig sensitized the uterus so that the proteose from the urine of this patient applied to the guinea pig uterus twenty-four hours later produced contraction. The serum from a patient sensitive to milk and using milk in the diet sensitized a guinea pig

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so that milk added to the perfusing fluid stimulated the uterus to contraction. Subsequently the patient's proteose gave a stronger contraction.

Boyd[4] found the ash content of proteose dried at 100 degrees C. to make up 70 to 87 per cent of the whole. It consisted of sodium sulphate with about 6 per cent calcium sulphate and some phosphate. The proteose contained 4.5 per cent nitrogen of which 3.2 per cent was soluble including 0.4 per cent ammonia nitrogen. Eighty-nine per cent of the material was soluble. The nitrogenous portion was found to be an irreversibly coagulable protein, possibly serum-albumin, and a mucoid containing 12.5 per cent nitrogen.

Subsequent reports from these and other observers have had to do largely with the clinical use of proteose.

Conybeare and Oriel[5] tested a group of pollen sensitive individuals with proteose extracted from pollen sensitive patients showing symptoms and concluded that since there were differences between the reactions of some patients to pollen extract and to proteose that the active substance in proteose was not identical to the original antigen.

Lyon, Percival and Stewart[6] found that a proteose from a hay fever patient which had been supplied them by Oriel gave nonspecific skin reactions. Working with proteose obtained from twenty-five patients they found four gave no positive reactions in any persons tested, fourteen were positive in all persons tested, one was positive in twenty-one of twenty-two, one in thirteen of twenty-two and five in one of two persons tested. Two proteoses from asthma-eczema patients gave negative reactions in the persons from whom they were derived but were positive in others. A proteose from a patient with angioneurotic edema was negative in the patient but positive in the skin of a patient with urticaria.

Freeman[7] also failed to secure results similar to those of the original workers. He found that skin reactions in hay fever patients were not specific and that he got positive reactions in normal persons. He stated that all proteose gave fairly constant toxic reactions appearing within a few minutes after intradermal injection. Reactions appearing from twelve hours to three days after injections were frequent but occurred as often in normal as in allergic persons. He believes that proteose is not a chemical substance but a more or less fortuitous substance produced by excretory activity of the kidney.

Minchin[8] reported the results of treatment of ten epileptics with proteose. He found no more skin reaction to the proteose than to dilute sodium hydrate. Three were treated with heterologous proteose with no improvement. Of seven treated with their own proteose four were unimproved and three showed temporary improvement.

Darley and Whitehead[9] in 1931 reported nine asthmatic persons examined. Three tested with proteose obtained in the interval between attacks gave no skin reaction. Two of these tested with proteose, obtained during attacks, reacted positively. Of four persons, negative to allergens usually used in testing, two were positive to their own proteose. Subcutaneous injection of proteose gave a focal reaction in four persons and desensitization with proteose gave good results in six of the nine. Two out of three persons suffering with serum sickness gave positive intradermal reactions to their own proteose. One chronic generalized eczema, one subsiding urticaria, and one erythema multiforme gave no skin reaction. Injection of proteose produced a leucocytosis and an eosinophilia. In 1932[10] they reported the treatment of twelve hay fever patients. To avoid severe reactions they used scratch tests and obtained definite wheals. Even focal reactions were said to occur. Of the twelve patients treated with their own proteose eight obtained complete, or almost complete relief, while four got partial relief.

Mills and Martyn[11] used proteose in the treatment of forty-four patients suffering with various chronic ocular diseases of obscure origin and claimed definite improvement in forty-one. In some of these they reported focal reactions from excessive dosage which they believed was conclusive evidence of the specificity of proteose.

Aldred-Brown and Munro[12] studied fifty cases of true active rheumatoid arthritis. Autogenous proteose was prepared from each. These patients were tested with intradermal and scratch, and a few with patch testing, and were found to be consistently negative to their own proteose. Six patients were treated with their own proteose, and in only one instance was any improvement obtained. The results were no better than those obtained by the ordinary methods of treatment. To test the toxicity of the proteose they injected one c.c. of the undiluted solution into the arms of fourteen patients. Eight gave no reaction at all, six gave slight local symptoms. They concluded that there was no specific activity of the proteose but called attention to the fact that it might have some nonspecific value in therapy.

Pearson[13] studied fifty asthmatic patients and fifty normal controls. He compared skin reactions to proteose with those produced by peptone, histamine and proteose. He found asthmatic persons giving nonspecific skin reactions to protein solution containing 10 to 20 milligrams of nitrogen per 100 c.c. This was less definite in the control group. Allowing for this tendency 38 per cent of the asthmatic patients gave significant reactions to their own proteose. In 24 per cent reactions occurred to the 1:100,000 dilution containing from 0.1 to 0.2 milligrams per 100 c.c. Of the asthmatic persons showing reaction to allergens, 50 per cent reacted to their own proteose while in those

showing no reaction to allergens only 15 per cent reacted to their proteose. Proteose from the asthmatic patients produced reactions in 32 per cent of the control group; 16 per cent reacted to a dilution of 1:100,000, and one half of these had an allergic family history. The skin reaction to histamine was found to be quite constant in both normal and asthmatic persons. They concluded that the skin reaction to proteose is due not to histamine but probably to some unidentified toxic substance to which the patient may be sensitized.

Eichenlaub,[14] investigating skin conditions, found three patients sensitive to their own proteose but not to others. He secured a positive Prausnitz-Küstner reaction in using the proteose from a patient with a neurodermatitis. Ten of eighteen patients gave positive skin reactions to their proteose and received benefit from treatment with the proteose. Eight gave negative skin reactions and only one of these benefited from treatment. Of five patients showing localized eczema one was cured by treatment with his proteose, three were improved and one was unimproved. Six patients with generalized eczema were similarly treated of whom three were markedly improved and three were failures. Of two persons with psoriasis one cleared completely while the other secured only temporary improvement. One neurodermite (also treated with x-ray) cleared up.

Trasoff and Meranze[15] working with asthmatic persons had disappointing results. Eight patients, all of whom reacted to certain allergens, showed very little reaction to their own proteose and reacted as well to heterologous proteose. No improvement from treatment with their own proteose was noted.

Cormia[16] found the average amount of proteose in 100 c.c. of urine of allergic persons to be 100 milligrams as contrasted with an average of 25 milligrams in normal persons. He tested five patients with eczema, five with urticaria, one with infantile eczema, three with chronic eczematous dermatitis, two with pruritus, one with dermatitis herpetiformis, two with asthma and three normal persons. He found no specificity in the reactions of any of these.

Vaughan[17] did skin tests on six patients with asthma, vasomotor rhinitis or dermatitis. He secured only borderline reactions to a 1:100 dilution of their proteose. Fourteen ragweed hay fever patients were tested. Five had symptoms when their proteose was obtained, and all were skin negative to the proteose. Of ten patients, all negative to their own proteose, seven gave reactions to ragweed and three were negative. He got no wheals on intracutaneous testing, never more than a flare. Two patients, treated with proteose, were unimproved.

Tuft and Brodsky[18] studied three groups of patients, one a control group of nonallergic individuals, another of patients suffering from nonallergic conditions, and the third group of patients presenting

symptoms of asthma, hay fever, urticaria and migraine. They found the amount of proteose put out by the kidney to vary rather widely, the largest amounts being found in the cardiovascular group. The allergic group eliminated somewhat larger amounts than the non-allergic controls and those suffering with hay fever produced somewhat more than the asthmatic patients. Skin tests done on twelve asthmatic patients were entirely negative in eight, very slightly reacted in four, but these four individuals also reacted similarly to normal proteose. Thirty patients suffering with hay fever were tested. Nineteen gave completely negative reactions, four slight reactions but similar reactions to the control, seven showed slightly larger reactions to the autogenous proteose than to the control but not sufficient difference to be considered specific. Treatment with autogenous proteose was carried out in three cases of long-standing asthma which had not yielded to other forms of treatment; no improvement was noted in either of these.

EXPERIMENTAL

The work to be reported may be divided into two parts. One, an effort to learn something of the identity of the substance in the urine responsible for whatever reactivity it may possess; two, a clinical investigation of the use of proteose in skin tests and in treatment.

The technic used in the preparation of proteose was that described by Oriel.[1] Except in the normal controls in every instance urine was secured while the patient was exhibiting symptoms of sensitization. The proteose obtained was used for testing within twenty-four hours of its preparation. Since Barber and Oriel found that ingestion of large amounts of glucose interfered with the activity of proteose, it was ruled out in the patients studied in our group.

Since it seems to be shown definitely that the amount of proteose is greatly increased by allergic reactions, no effort was made to determine the amounts in twenty-four-hour output in allergic and normal persons.

The Millon, xanthoproteic and biuret reactions were positive. The Molisch reaction was invariably present in the specimens tested. By the method of Rimington[19] a small amount of a complex carbohydrate could be recovered. The larger part of the material consisted of inorganic salts. Nitrogen determinations made on six specimens showed 4.4 per cent nitrogen of which 0.38 per cent was ammonia nitrogen. Precipitation by ammonium sulphate was almost complete. There seemed to be practically no difference in the character of the material though derived from various persons suffering with different types of sensitization.

In the belief that proteose owes its activity to unchanged allergens absorbed into the blood stream and eliminated in the urine, tests were

made on three grass sensitive and three ragweed sensitive patients all of whom were showing active hay fever symptoms at the time of examination. The proteose from two of these sensitive to grass and all three of the ragweed sensitive patients produced definite wheals with a zone of hyperemia when injected intradermally into the patients from whom derived. Questionable or no reactions were obtained by scratch tests. When injected into members of this group, other than the one from whom derived, the reaction was found to vary as did the reaction of the respective individuals to dilution of grass or ragweed pollen. When compared in the skin of these persons with different concentrations of grass and ragweed pollen, it was found that the proteose behaved as a weak dilution of the pollen extract. Table I illustrates these findings. It will be seen that patient B re-

TABLE I

SKIN REACTIONS TO PROTEOSE

	DILUTION	500	2,000	5,000	20,000	50,000	100,000
Patient A	Ragweed	+++	++	++	+	±	−
	Proteose A	+	±	−	−	−	−
	Proteose B	+	+	−	−	−	−
	Proteose C	±	−	−	−	−	−
Patient B	Ragweed	+++	+++	++	+	+	+
	Proteose A	++	+	+	−	−	−
	Proteose B	++	++	+	+	−	−
	Proteose C	+	±	−	−	−	−
Patient C	Ragweed	+	+	+	±	−	−
	Proteose A	±	−	−	−	−	−
	Proteose B	+	±	−	−	−	−
	Proteose C	±	−	−	−	−	−

acted to higher dilutions of ragweed than the other two while patient C reacted the least. B likewise reacted to all three proteoses more than the others and C the least. It may be seen that patient B reacted to his own proteose more strongly than did the others, and he likewise reacted to proteose A and C more markedly than the patients from whom they were derived. Likewise patient C reacted more to the proteose from B and least to his own. This would seem to indicate that B, who reacted most to ragweed pollen, reacted most to all three proteoses and the others reacted to proteose in somewhat the same manner, though to a much less degree, as to the pollen. It would seem to indicate, also, that B eliminated in his urine more of the active substance than did the others. These findings may be construed as evidence that the reaction to proteose varies with the reactivity of the skin to the allergen to which the patient is sensitive and that the amount of active substance in proteose may vary considerably in different persons.

In a previous communication[20] on the subject of the oral administration of pollen it was shown that pollen taken by mouth is eliminated

to a demonstrable extent in the urine. Taking advantage of this fact, ragweed pollen was administered orally to one of us (B. S.) for five days beginning with five drops of the 1:50 dilution three times daily and increased by five drops three times a day each day until twenty-five drops were being taken three times a day on the fifth day. Proteose was extracted from the urine each day for six days, beginning with the first day and continuing through the day following the last injection of pollen. Urine secured the first day was used as a control, and the proteose was found to give no reaction in skin test in ragweed sensitive persons. Proteose obtained on each of the following five days gave definite skin reaction in ragweed sensitive persons, but not in B. S. who is not ragweed sensitive and whose proteose it was. Forty ragweed sensitive patients and forty not sensitive to ragweed were tested with these proteoses pooled. The results of skin tests on ten ragweed sensitive patients and two non-sensitive persons, illustrating results

TABLE II

SKIN REACTIONS ON RAGWEED SENSITIVE INDIVIDUALS

PERSON TESTED	B. S PROTEOSE	J. H. B. PROTEOSE	RAGWEED 1:500
B. S.	−	−	−
J. H. B.	+	−	++
A	++	−	+++
B	+	−	++
C	+	−	++
D	±	−	++
E	+	−	++
F	+	−	+
G	+	−	+++
H	−	−	+
I	−	−	+
Control	−	−	−
Control	−	−	−

obtained, are shown in Table II. Here the reactions are compared with the reactions to ragweed pollen extract (intradermal injection of 1:500) and to a proteose obtained from the urine of J. H. B. who is ragweed sensitive but was not exposed to ragweed pollen at the time and was showing no symptoms. Different dilutions of proteose were not used. The number of pluses shows the intensity of reaction to 1:100 dilution. It may be seen that B. S., who is not sensitive to ragweed, did not react to his own proteose, but eight out of ten ragweed sensitive persons did. The two negative reactions were in those who had slight reactions to ragweed. There were no reactions in the normal controls. There were no reactions to the proteose of J. H. B. Of the forty ragweed sensitive persons tested thirty-three reacted to B. S's proteose. These findings may permit the conclusion that ragweed pollen, eliminated in the urine, is extracted with the proteose and that this proteose behaves as does that obtained from a pollen sensitive

patient showing active symptoms. In the experiment the urine was not secreted during an attack of hay fever and the elimination of the ragweed pollen and proteose occurred independent of any allergic mechanism.

It is possibly worthy of mention that the amount of proteose eliminated by B. S. did not vary appreciably in the six specimens obtained. This would seem to indicate that the marked increase in elimination of proteose during an allergic attack may be due to some factor of the attack which may be independent of the amount of antigen eliminated.

CLINICAL

In addition to the tests made on pollen sensitive persons, as described above, thirty-seven patients were tested with their own proteose and four received treatment with it. Four normal persons were used as controls. Two patients suffered with perennial hay fever and four with asthma. The others were all manifesting some skin lesions, and the proteose was secured during activity of the condition. A few of these are not regarded generally as allergic. The four normal persons gave no reactions to their own proteose, but three of them reacted to one or more proteoses from patients.

Two perennial hay fever patients, sensitive to foods, did not react to their proteose. Of the four asthmatic persons, three did not give any skin reaction to any allergen and their etiology was unknown. They gave no reaction to proteose. One was given five injections of his proteose without reaction and without improvement. The fourth was pollen sensitive and reacted to his own proteose. One patient with pruritus of unknown origin did not react to his proteose. One angioneurotic edema patient, etiology unknown, reacted to his proteose but so did three of the four controls. Twenty-one injections of proteose did not bring about any improvement in his condition.

Five patients with urticaria were tested. One of these apparently had its origin in a sensitization to tricophyton; the other four were of unknown etiology. The patient with tricophyton infection and one other reacted to their proteose while the controls were negative. One patient who did not react and one who did (not trichophyton sensitive) were treated with their proteose, one receiving eight and the other fifteen injections without benefit.

One patient with a phenolphthalein eruption was negative to her proteose. She had none of the drug for several days before securing the proteose but had a widespread eruption at the time.

Three persons with eczema due to food were negative. One of these, sensitive to wheat, had eaten wheat and produced an acute eczema at the time the urine was secured. The others were chronic conditions.

Two seborrheic eczemas gave negative reactions. One patient showing dermographism was negative. One patient with severe trichophyton infection of both feet was negative. Four patients with contact eczemas were all negative to their proteose. Of ten persons with generalized eczema two reacted to their own proteose but so did normal controls. One patient with a diagnosis of dysidrosis was negative and one diagnosed as a neurodermite was also negative.

It should be stated that reactions in these patients with skin lesions did not consist of the irregular wheal with a zone of hyperemia which might be found in the pollen cases but showed a small wheal or indurated area with surrounding hyperemia and often appeared four or more hours after the injection.

SUMMARY

There seems to be sufficient evidence to suggest that the activity and specificity of proteose may be due to unchanged antigen which has been eliminated by the kidneys and may be carried along with the proteose in its extraction from the urine. Reactions may be secured fairly regularly in pollen sensitive patients who give definite reactions to pollen. Reactions are rarely secured in those sensitive to other allergens which is analogous to the common finding of fewer and less marked reactions to allergens in this group than to pollen.

The results of testing with proteose from a non-sensitive person to whom pollen was given orally would show that pollen is extracted from the urine by the method of proteose extraction and the proteose from this individual behaved as a weak solution of pollen. The fact that this proteose was not increased in amount by the ingestion of pollen as occurs in allergic attacks may indicate that the allergic reaction produces an increased elimination of the proteose which may be entirely independent of the amount of antigen eliminated with it.

It would not seem necessary to hypothecate a new substance or an antigen-tissue factor combination to explain the results obtained.

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CAMOMILE *(ANTHEMIS COTULA)* AS A SKIN IRRITANT

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DURING the last year and a half my studies have shown that camomile *(Anthemis cotula)* is a common skin irritant and as such may be a cause of dermatitis venenata. The reacting substance in it is similar to that of sumac, poison oak, poison ivy,[1] orange peel,[2] crysanthemum,[3] burweed, marsh elder,[4] the heleniums,[5] tansy,[6] common bleeding heart,[7] pollen lipoids or oils,[8] and other substances which owe their production of dermatitis to an oil or resin. The only previous reference to camomile as an irritant to the skin is its inclusion in the extensive list of skin irritants published by Weber.[9]

Attention to this skin irritant was the result of study of a patient referred to me on May 23, 1932, by Dr. E. Chipman of San Francisco. The man, aged sixty-two years, with a negative personal and family history of allergy had worked on a farm in Monterey County for sixteen years cultivating and cutting alfalfa and driving horses. He had no contact with chickens, cows or other animals. In June, nine years ago, he first noticed an itching rash on his face, and since then this had gradually spread over his neck, hands, wrists, scrotum, backs of knees, legs and ankles. He had never been free though the dermatitis was much diminished in the winters and when he remained indoors and especially if he stayed in San Francisco. During June, July and August, it became so diffuse, itching and severe that he was unable to continue his routine work. Tests with over 200 food, animal emanation, pollen, dust, grain and miscellaneous allergens were negative with the scratch test. With the intradermal test one-plus reactions occurred to *Cynodon dactylon, Chenopodium album, Artemesia californica, Ambrosia psilostachya, Franseria bipinnatifida,* wheat dust, rice sack dust, barley dust, and two-plus reactions to oat dust and alfalfa dust. The patient was then patch tested with grain feeds, animal emanations, various substances encountered in his living environment and finally with leaves and flowers of all vegetation with which he came in contact in his garden, in the fields and on the hills. All these were negative except for a violent vesicular itching reaction to camomile *(Anthemis cotula)* which became definite in twelve hours. This oozing lesion required a dressing for one week. Scratch test with an extract of this weed gave no immediate wheal reaction.

He was injected with one extract containing *Chenopodium album, Atriplex patula, Artemesia californica* and *Ambrosia psilostachya* and

another containing alfalfa, *Bromus carinatus* and *Lolium perenne* during the fall months. He also took a strong watery extract of the indigenous camomile by mouth. His first dose was two teaspoonfuls, which produced marked swelling and irritation of his throat and burning of his stomach in about one-half hour, making swallowing difficult

Fig. 1.—Camomile (*Anthemis cotula*).

Fig. 2.—Camomile (*Anthemis cotula*).

for many hours. He then began with a drop dose and gradually was able to increase it over a period of three months up to two cupfuls without local reactions. He continued to take a cupful of this tea daily through the spring months. At present his patch reaction to camomile is still positive.

The lesions gradually disappeared completely during the fall and winter, and no eruption occurred at the usual time of onset from May 15 to June 1. On July 21, however, after working hard on a hot day in the alfalfa fields he had a mild outbreak on his face, arms and calves of legs. He was relieved in a few days on coming to San Francisco. Camomile tea, which had been discontinued one month previously, was begun again, and an extract containing alfalfa, *Lolium perenne, Chenopodium album, Artemesia vulgaris* and *Atriplex patula* was again administered. He returned to work and has had no further eruption in the last six months.

This patient was abnormally sensitive to camomile as evidenced in the mucous membrane reaction and the marked skin reactions to the

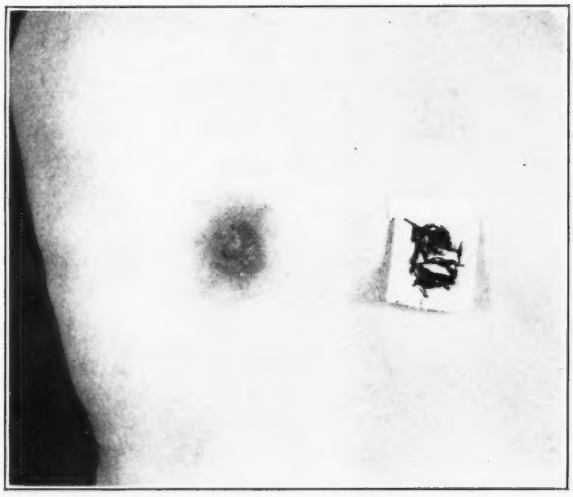

Fig. 3.—The twenty-four-hour reaction from patch test with camomile (*Anthemis cotula*).

patch test. It seems probable that the oral administration of the camomile extract was at least partly responsible for the marked delay in reappearance of the lesions. The desensitization to alkalfa, weed and grass pollens probably was helpful. However, his pollen reactions were very small, and he has daily contact with grain, and alfalfa dusts when feeding and milking cows every winter when his dermatitis previously has always greatly diminished.

That *Anthemis cotula* may be nearly a universal irritant to the skin is evident through the following experiments. Patch tests with dry *Anthemis cotula* on the normal skin of 21 adults not suffering with skin lesions have given in 16 cases definite areas of dermatitis, as shown in Table I, requiring days to heal, and remaining irritated and pigmented for several weeks. Only two persons failed to give a twenty-four-hour or delayed reaction. All were likewise tested with *Anthemis nobilis*

(Roman camomile) a commercial product as purchased in the drug store, and only 4 of the 21 subjects gave slight or questionable reactions. The patients who have a marked dermatitis from camomile probably have a constitutional predisposition which makes their skin hypersusceptible to the resin of this plant. Twelve other normal people were tested with *Anthemis cotula* and *Matricaria chamomilla* (German camomile) as purchased from the druggist. All gave marked twenty-four-hour and delayed reactions to the first species and none reacted to the second. The fewer reactions to the two commercial products may be due to the excessive dryness of the product. It may indicate, however, that the skin susceptibility is species specific for the local indigenous comomile. Through the courtesy of Dr. Francis Scott Smyth patch tests with *Anthemis cotula* on seven infants not more than four days old were made and have yielded no twenty-four-hour or delayed reactions. This indicates that skin sensitivity to this weed probably requires previous contact such as seems necessary for establishment of sensitization to poison ivy as demonstrated by Straus.[10] Further tests should be made on adults who live in regions where *Anthemis cotula* never grows.

TABLE I

RESULTS OF PATCH TESTING WITH TWO SPECIES OF CAMOMILE

	WITHIN 24 HOURS					DELAYED			
	0	±	+	++	NO READING	0	+	++	+++
Anthemis cotula (Indigenous)	2	3	11	5	2	2	1	14	2
Anthemis nobilis (Roman camomile)	17	1	3		2	17		2	

Extract of the dried leaves and flowers of *Anthemis cotula* made with water, 20 per cent ethyl alcohol, and normal saline solution gave definite reactions in twenty-four hours by the patch test on four individuals. The irritating substance, however, was completely removed by extracting the leaves and flowers with fresh ether for two hours, ten different times. The first four ether extractions were saved and evaporated. This resinous residue gave positive patch tests on the skin of three normal individuals. Recently, tests have been done with the young leaves of camomile *(Anthemis cotula)* before flowering had occurred and positive reactions have not been definite. This may indicate that the reacting substance does not form in the weed until maturity has occurred. The distribution of *Anthemis cotula* throughout the United States has been ascertained by questionnaires sent to botanical departments in colleges and universities in every state. Chart 1 shows it is in every state except Florida, occurring especially in fields, along roads, in vacant lots, barnyards, henyards, homesteads, uncultivated

grounds and waste places. Other related species are frequently found especially in eastern states. It is absent in the Hawaiian Islands.

In his answer to my questionnaire, Professor L. Donald of Mississippi wrote: "I think I voice the sentiment of landowners throughout the state in saying that this plant is one of the most obnoxious weeds occurring in pastures, meadows and hay fields. Considerable trouble is also incurred by its presence among crop products, especially among the small grains." Professor P. C. Standley of Chicago stated: "I remember that the odor of the plant was seldom absent when driving along countrysides. My grandmother was very sensitive to the plant, and it was almost as dangerous to her as was poison ivy! In addition a patient stated that in western Washington, where camomile grows

Chart 1.—Distribution of *Anthemis cotula* in the United States.

quite extensively, many people had a rash on their legs after walking through camomile wet with the dew or rain."

This report directs attention to camomile *(Anthemis cotula)* as a nearly universal skin irritant here in California and probably in other regions where the weed grows in any abundance. The fact that newborn infants do not react may indicate that skin reactivity requires previous sensitization by actual exposure to the weed. Failure to react to the European varieties may be due to the necessity of previous exposure or sensitization to the specific species, but this is being given continued study. It is probable that the skin of certain individuals is so sensitive to the resinous emanations of this weed that the amount in the air and no actual contact with the weed is sufficient to establish a severe and at times a progressively diffuse dermatitis. Such exposure could occur by merely riding through the countryside.

CONCLUSIONS

Anthemis cotula produces definite vesicular erythematous skin lesions within twenty-four hours in practically all children and adults with the patch test. Infants in the first days of life do not react. Certain patients are abnormally hypersensitive to this weed and may develop a dermatitis venenata of varying severity. The specific reacting substance is an oil or resin.

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SURVEY OF A BOTANIC OASIS IN THE DESERT OF NORTHERN ARIZONA*

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IN THE north central portion of Arizona is a plateau 225 miles in extent, practically all of which is at an altitude of 5,000 feet. Arising in about the center of this broad expanse is another plateau, the San Franciscan Volcanic Field. Much of this is fairly level and has an altitude of 6,500 to 7,000 feet. This volcanic field is abruptly ter-

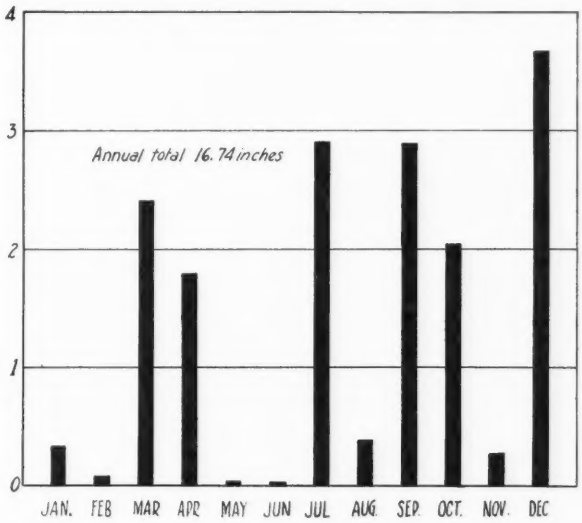

Fig. 1.—Monthly and total rainfall for Flagstaff, Arizona, during 1924.

minated on the west and south by the precipitous Aubrey Cliffs. Bill Williams Mountain, the largest extinct volcano in the region, rises to a height of 9,600 feet in the western half of this area. To the east are the San Francisco Peaks, with an altitude of nearly 13,000 feet, the highest point in Arizona. The towns of Williams, on the west, and Flagstaff, at the foot of the San Francisco Peaks to the east, are within the area of this volcanic uplift.

The climate of this region is typical of that of an arid to semiarid section. The temperature and rainfall of Flagstaff for each of two years are indicated in Figs. 1, 2, 3 and 4. The chief characteristic of the rainfall is its extreme variability. In general there are two wet seasons, one in summer and the other in winter, separated by much

*From the Allergy Clinic, Santa Fe Coast Lines Hospital Association.

drier periods. In the winter snow falls in most of this section. At Williams the annual average rainfall amounts to 15 inches; and at Flagstaff, 66 miles east, it is 22 inches, the greatest found at any sta-

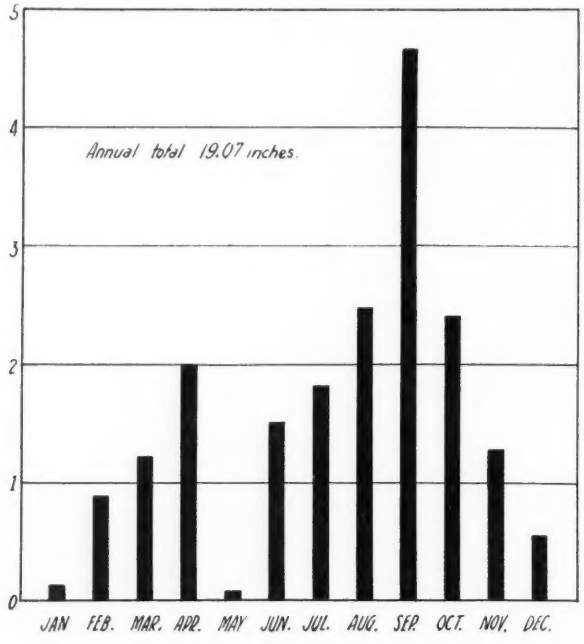

Fig. 2.—Monthly and total rainfall of Flagstaff, Arizona, during 1925.

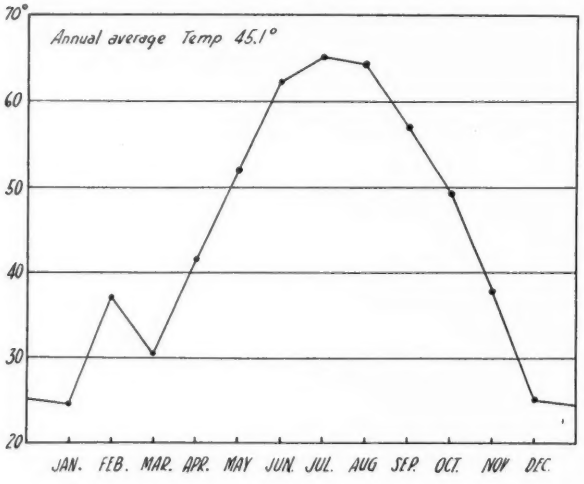

Fig. 3.—Monthly and annual average temperatures for Flagstaff, Arizona, during 1924.

tion in Arizona. The precipitation at the latter point is influenced, no doubt, by the proximity of the San Francisco Peaks.

Although the Williams-Flagstaff area is encircled on all sides by a barren and arid region, much of which is in fact a desert, it is clothed

in a beautiful forest of juniper, pine and spruce. The abrupt change from desert to forest is most strikingly seen in passing from Winslow westward toward this section. This is emphasized in Fig. 5.

The botanic survey of these two communities was made in 1930-31. The methods employed have been outlined in previous reports[1-3] and need not be discussed here. The detailed findings at the several sampling stations, from west to east, will be indicated in subsequent paragraphs.

WILLIAMS, ARIZONA

Station 1, which is located two miles west of the center of town on highway 66, is just at the entrance to the Mountain Spring Ranch and comprises an area on either side of the road. The dominating plant of this montane forest section is *Pinus ponderosa*. Of the grasses,

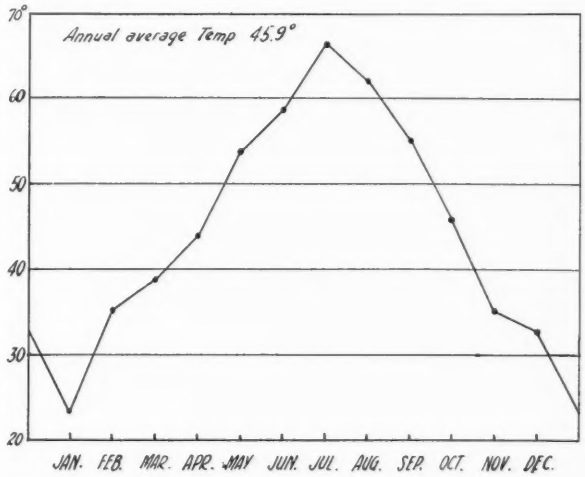

Fig. 4.—Monthly and annual average temperatures for Flagstaff, Arizona, during 1925.

which are common subdominants, *Bouteloua hirsuta* and *Sitanion hystrix* are the most abundant. The Chenopodiaceae is represented by *Chenopodium album* and *Salsola pestifer*, especially abundant along the highway. *Gutierrezia sarothrae* is the most abundant Compositae, but *Artemisia camporum*, *Ambrosia psilostachya*, *Helianthus annuus* and *Solidago glaberrima* are quite common.

Station 2 is located halfway between Station 1 and town. Like the previous station it is a *Pinus ponderosa* consociation and presents a similar picture of subdominants except that *Aristida fendleriana*, *Agropyron tenerum*, and *Bouteloua gracilis* are here quite numerous.

Station 3, two miles south of Williams, is in a canyon of Bill Williams Mountain, at the head of the cataract. The higher elevation probably accounts for the occurrence of a *Pinus ponderosa-Pseudotsuga mucronata* association, with *Populus aurea*, "Quaking aspen," rather common

TABLE I

BOTANIC SURVEY OF WILLIAMS, ARIZONA

SCIENTIFIC NAME	COMMON NAME	STATIONS 1	2	3	4	5	6	SEASON OF POLLINATION
PINACEAE	PINE FAMILY							
Pinus ponderosa	Yellow pine	****	****	***	***	**	–	El. Sp.
Pseudotsuga mucronata	Douglas spruce	–	–	**	–	–	**	El. Sp.
CUPRESSACEAE	CYPRESS FAMILY							
Cupressus sp.	Cypress	**	*	–	–	**	–	El. Sp.
Juniperus monosperma	One-seeded juniper	–	**	–	–	–	–	El. Sp.
GRAMINEAE	GRASS FAMILY							
Agropyron tenerum	Slender wheat grass	***	***	–	–	***	***	Sp. Sm.
Agrostis hiemalis	Hair grass	**	–	**	–	–	–	Sp.
Agrostis palustris	Redtop	–	–	**	–	–	–	Sm.
Aristida fendleriana	Fendler's aristida	–	***	–	***	**	–	Sp. Sm.
Aristida longiseta	Wiregrass	–	–	–	***	–	–	Sp. Sm.
Bouteloua gracilis	Blue grama	–	***	–	***	**	****	Sp.
Bouteloua hirsuta	Black grama	***	**	–	–	****	–	Sm.
Bouteloua procumbens	Six weeks grama	*	–	–	–	–	–	Sm.
Bromus commutatus	Downy-sheathed cheat	**	–	–	–	–	–	Sm.
Bromus marginatus	Large mountain brome grass	–	–	**	–	–	–	Sp. Sm.
Bromus polyanthus paniculatus	Great basin brome grass	***	–	–	–	–	–	Sm.
Bromus tectorum	Downy brome	–	–	–	–	–	**	Sp.
Echinochloa crusgalli	Barnyard grass	–	–	*	**	–	**	Sp.
Epicampes rigens	Deer grass	**	–	–	–	–	–	Sp. Sm.
Eragrostis mexicana	Mexican eragrostis	–	**	–	**	–	–	Sp.
Hordeum jubatum	Squirrel tail	–	–	–	–	***	***	Sp.
Muhlenbergia filiformis	Slender muhlenbergia	–	–	***	–	–	–	Sp. Sm.
Muhlenbergia squarrosa		–	–	–	–	–	***	Sp. Sm.
Panicum capillare	Old-witch grass	–	–	–	*	–	–	Sm.
Phleum pratense	Timothy	–	–	**	–	–	–	Sm.
Poa bigelovii (identification uncertain)		–	–	–	–	–	**	Sp. Sm.
Poa compressa	English blue grass	–	–	**	–	–	–	Sp.
Sitanion hystrix	Bottle-brush squirrel tail	***	***	***	**	–	–	Sp. Sm.
Triodia pulchella	Low triodia	*	–	–	–	–	–	Sm.

TABLE I—CONT'D

SALICACEAE	WILLOW FAMILY							
Populus aurea	Quaking aspen	–	–	***	–	–	–	El. Sp.
Populus alba	White poplar	–	–	–	–	–	**	El. Sp.
Populus fremontii	Fremont cottonwood	–	–	–	–	–	**	El. Sp.
Salix irrorata	Willow	**	*	–	–	–	–	El. Sp.
FAGACEAE	OAK FAMILY							
Quercus sp.	Oak	**	*	**	–	–	–	Sp.
POLYGONACEAE	BUCKWHEAT FAMILY							
Eriogonum racemosum	Buckwheat	**	–	–	–	–	–	Sm.
Polygonum sp.	Knotweed	**	**	–	–	**	–	Sm.
Rumex acetosella	Sheep sorrel	–	–	*	–	–	–	Sm.
Rumex crispus	Curly dock	**	**	**	–	–	–	Sm.
CHENOPODIACEAE	SALTBUSH FAMILY							
Atriplex rosea	Red orache	–	–	–	–	****	***	Sm. Fl.
Chenopodium album	Lamb's-quarters	****	***	***	–	***	****	Sm.
Kochia scoparia	Firebush or Summer cypress	–	–	–	–	–	**	Sm.
Salsola pestifer	Russian thistle	***	***	–	***	–	****	Sm. Fl.
AMARANTHACEAE	PIGWEED FAMILY							
Amaranthus retroflexus	Rough pigweed	**	–	**	–	**	**	Sm. Fl.
RANUNCULACEAE	BUTTERCUP FAMILY							
Thalictrum fendleri	Meadow rue	–	**	–	–	–	–	Sm.
CRUCIFERAE	MUSTARD FAMILY							
Thelypodium wrightii		****	–	–	–	–	–	Sm.
ROSACEAE	ROSE FAMILY							
Cowania stansburiana	Cliff rose	–	–	–	**	–	–	Sm.
Rosa helleri	Rose	*	–	*	–	–	–	Sp. Sm.

TABLE I—CONT'D

SCIENTIFIC NAME	COMMON NAME	STATIONS 1	2	3	4	5	6	SEASON OF POLLINATION
LEGUMINOSAE	PEA FAMILY							
Lotus trispermus	Bird's-foot trefoil	**	–	*	–	–	–	Sm.
Lupinus kingii	Lupine	–	–	**	–	–	–	Sm.
Melilotus alba	White melilot or sweet clover	*	–	–	–	**	–	Sm.
Psoralea tenuiflora		**	–	–	–	–	–	Sm.
Robinia neomexicana	New Mexican locust	**	–	–	–	–	–	Sp.
Trifolium sp.		–	–	*	–	–	–	Sm.
GERANIACEAE	GERANIUM FAMILY							
Geranium furcatum	Cranesbill	***	***	–	–	***	***	Sm.
EUPHORBIACEAE	SPURGE FAMILY							
Euphorbia sp.	Spurge	–	–	–	–	–	*	Sm.
ACERACEAE	MAPLE FAMILY							
Acer sp.	Maple	–	–	-	–	–	*	Sp.
Acer negundo	Box elder	–	–	–	–	–	**	Sp.
RHAMNACEAE	BUCKTHORN FAMILY							
Ceanothus sp.		–	–	**	–	–	–	Sp.
MALVACEAE	MALLOW FAMILY							
Malva crispa	Curled mallow	–	–	–	–	–	*	Sm.
Sidalcea neomexicana		***	–	–	–	–	**	Sm.
TAMARICACEAE	TAMARISK FAMILY							
Tamarix gallica	French Tamarisk	–	–	–	–	–	**	Sm.
ASCLEPIADACEAE	MILKWEED FAMILY							
Asclepias mexicana	Narrow-leaf milkweed	–	–	–	–	–	*	Sm.
CONVOLVULACEAE	MORNING GLORY FAMILY							
Convolvulus arvensis	Bindweed	**	–	–	–	–	–	Sm.
POLEMONIACEAE	GILIA FAMILY							
Gilia aggregata	Scarlet gilia	***	–	**	–	–	–	Sm.
Gilia pumila		–	–	–	**	–	–	Sm.

TABLE I—CONT'D

LABIATAE	MINT FAMILY							
Monarda menthaefolia	Horsemint	–	–	**	–	***	–	Sm.
SCROPHULARIACEAE	FIGWORT FAMILY							
Castilleia sp.	Painted cup	–	–	*	–	–	–	Sm.
Verbascum thapsus	Common mullein	**	–	–	–	–	–	Sm.
PLANTAGINACEAE	PLANTAGO FAMILY							
Plantago argyrea	Plantain	***	–	–	–	–	–	Sm.
COMPOSITAE	SUNFLOWER FAMILY							
Achillea lanulosa	Yarrow	**	–	*	–	–	–	Sm.
Ambrosia psilostachya	Western ragweed	***	**	–	***	**	**	Fl.
Artemisia camporum		***	***	****	**	**	–	Fl.
Artemisia carruthii		**	***	–	–	–	–	Fl.
Aster sp.		–	–	*	–	–	–	Sm.
Cirsium wheeleri	Thistle	*	–	–	–	–	–	Sm.
Chrysothamnus graveolens	Rabbit brush	**	–	–	–	**	–	El. Sm.
Dieranocarpus dieranocarpus		*	–	–	–	–	–	Sm.
Erigeron sp.		*	–	–	–	–	–	Sm.
Franseria discolor		**	–	–	–	–	**	Fl.
Grindelia aphanactis	(identification uncertain)	–	**	–	–	***	–	Fl.
Gutierrezia sarothrae	Matchweed	****	****	–	***	****	***	Fl.
Haplopappus gracilis		*	–	–	–	–	–	Sm.
Helianthus annuus	Common sunflower	***	–	–	–	–	***	Sm. Fl.
Lactuca scariola	Prickly lettuce	**	–	–	–	–	–	Sm. Fl.
Sanvitalia aberti		–	–	**	–	–	–	Fl.
Senecio sp.	Groundsel	*	–	–	–	–	–	Sm. Fl.
Solidago glaberrima	Goldenrod	***	–	**	–	–	–	Sm.
Taraxacum taraxacum	Dandelion	*	–	–	–	–	–	Sm.
Verbesina encelioides exauriculata	Crown-beard	–	–	–	–	–	**	Sm.

Controls—All sites negative.
* = present or rare.
** = fairly common, few.
*** = common.
**** = very common or abundant.

– = does not occur.
Fl. = pollinates in the fall.
Sp. = pollinates in the spring.
Sm. = pollinates in the summer.
El. = early in the season indicated.

along the streams. *Sitanion hystrix* and *Muhlenbergia filiformis* are the only grasses of note. Again *Chenopodium album* is common and *Artemisia camporum* is quite abundant.

Station 4 is near the mouth of the same canyon by the Sante Fe Dam at a distance of one-half mile from center of town. *Agrostis fendleriana, A. longiseta* and especially *Bouteloua gracilis* are nearly equal in importance to the dominant, *Pinus ponderosa. Salsola pestifer* is the common member of the Chenopodiaceae and *Ambrosia psilostachya* and *Gutierrezia sarothrae* are the most significant Compositae.

Station 5, one mile north of the highway, is along the old road to Grand Canyon. Between this station and the highway is an expanse of cleared field, and only a very few *Pinus ponderosa* are present. The most important plants are *Gutierrezia sarothrae, Atriplex rosea* and *Bouteloua hirsuta,* which are found abundantly; and *Hordeum jubatum, Agropyron tenerum* and *Chenopodium album* are somewhat less common.

Station 6 includes a relatively large area and is in reality a composite of many small stations scattered throughout the entire residential section. A number of ornamental trees such as *Populus alba, P. fremontii, Acer negundo* and *Tamarix gallica* are cultivated. Perhaps the only shrub of any importance, *Kochia scoparia,* should be mentioned. There are but few lawns, and no *Cynodon dactylon* was collected from this town. Thus, practically the only significant grasses are the native ones, such as *Agropyron tenerum, Bouteloua gracilis, Hordeum jubatum* and *Muhlenbergia squarrosa.* The Chenopodiaceae quite common at this station are *Chenopodium album, Atriplex rosea,* and *Salsola pestifer. Gutierrezia sarothrae* and *Helianthus annuus* are also rather abundant.

FLAGSTAFF, ARIZONA

Station 1 is situated along the highway about two miles west from the center of the town. At this point we find the characteristic *Pinus ponderosa* consociation and both *Agropyron tenerum* and *Bouteloua gracilis* equal *Sitanion hystrix* in abundance. *Chenopodium album* is the only significant representative of the Chenopodiaceae. *Artemisia camporum, Franseria tenuifolia* and *Helianthus annuus* are all rather common.

Station 2, just on the western edge of the residential section, is characterized by the dominance of Chenopodiaceae; most prominent of which are *Salsola pestifer, Atriplex rosea* and *Chenopodium album. Ambrosia psilostachya* is more common here than at the previous station, and *Sitanion hystrix* appears as the most significant grass present. The pines have, of course, been removed from this section.

Station 3 is just south of the center of town and beside the railroad station. A number of grasses, such as *Dactylis glomerata, Festuca arizonica, Hordeum jubatum, Lolium perenne* and *Poa pratensis* are

TABLE II

BOTANIC SUMMARY OF FLAGSTAFF, ARIZONA

SCIENTIFIC NAME	COMMON NAME	STATIONS 1	2	3	4	5	6	SEASON OF POLLINATION
PINACEAE	PINE FAMILY							
Pinus ponderosa	Yellow pine	***	–	–	–	–	****	El. Sp.
Juniperus monosperma	One-seeded juniper	***	–	–	–	–	***	El. Sp.
GRAMINEAE	GRASS FAMILY							
Agropyron caninum	Bearded wheat grass	–	–	–	–	**	–	Sp.
Agropyron repens	Quack grass	–	–	–	–	**	–	Sp.
Agropyron richardsoni		–	–	–	–	**	–	Sp.
Agropyron smithii var. *molle*	Western wheat grass	–	–	–	–	–	**	Sp.
Agropyron tenerum	Slender wheat grass	***	**	**	–	****	***	Sm.
Aristida purpurea	Purple aristida	–	**	–	–	–	–	Sm.
Bouteloua gracilis	Blue grama	***	–	–	–	***	**	Sp. Sm.
Bromus polyanthus		–	*	–	–	**	–	Sp. Sm.
Bromus tectorum	Downy brome	–	–	**	–	**	–	Sm.
Dactylis glomerata	Orchard grass	–	–	***	–	*	–	Sp. Sm.
Epicampes rigens	Deer grass	**	–	–	–	**	–	Sp.
Eragrostis mexicana	Mexican eragrostis	–	–	**	–	–	–	Sm.
Festuca arizonica	Arizona fescue	**	–	***	–	–	–	Sm.
Hordeum jubatum	Squirrel tail	–	**	***	–	**	**	Sp. Sm.
Lolium perenne	Perennial rye grass	–	–	***	–	**	–	Sp.
Muhlenbergia montana	Mountain muhlenbergia	**	–	–	***	–	–	Sm.
Poa annua	Annual blue grass	–	–	–	–	**	–	Sm.
Poa pratensis	Kentucky bluegrass	–	–	***	**	–	***	Sp.
Poa scabrella	Malpais blue grass	–	–	–	–	**	–	Sp.
Schedonnardus paniculatus	Texan crab grass	*	–	–	–	–	–	
Sitanion hystrix	Bottle-brush squirrel tail	***	***	–	–	**	**	Sp. Sm.
Triodia pulchella	Low triodia	**	–	–	–	–	–	Sp. Sm.
SALICACEAE	WILLOW FAMILY							
Populus alba	White poplar	–	–	–	*	–	–	El. Sp.
Populus fremontii	Fremont cottonwood	–	–	**	**	–	–	El. Sp.

TABLE II—CONT'D

SCIENTIFIC NAME	COMMON NAME	STATIONS 1	2	3	4	5	6	SEASON OF POLLINATION
POLYGONACEAE	BUCKWHEAT FAMILY							
Eriogonum alatum	Buckwheat	*	–	–	–	*	–	Sm.
Rumex sp.		–	–	–	*	*	–	Sm.
CHENOPODIACEAE	SALTBUSH FAMILY							
Atriplex rosea	Red orache	**	***	***	***	–	**	Sm. Fl.
Chenopodium album	Lamb's-quarters	***	***	****	**	***	**	Sm. Fl.
Chenopodium leptophyllum		–	–	**	–	*	–	Sm.
Salsola pestifer	Russian thistle	–	****	****	**	**	**	Sm.
AMARANTHACEAE	AMARANTH FAMILY							
Amaranthus retroflexus	Rough pigweed	–	**	***	–	**	–	Sm. Fl.
RANUNCULACEAE	BUTTERCUP FAMILY							
Thalictrum fendleri	Meadow rue	**	–	–	–	–	–	Sm.
CAPPARIDACEAE	CAPER FAMILY							
Cleome serrulata	Rocky Mountain bee plant	–	**	**	–	–	–	Sm.
CRUCIFERAE	MUSTARD FAMILY							
Thelypodium wrightii		**	–	–	–	–	–	
LEGUMINOSAE	PEA FAMILY							
Acuan jamesii		–	–	–	–	–	**	Sm.
Lotus tripermus	Trefoil	**	–	–	–	–	–	Sm.
Lupinus kingii	Lupine	*	–	–	–	–	–	Sp.
Parosela jamesii		–	–	–	–	**	–	Sp.
Robinia pseudacacia	Black locust	–	–	**	–	–	–	Sp.
GERANIACEAE	GERANIUM FAMILY							
Geranium furcatum	Cranesbill	***	–	–	–	–	**	Sm.
MALVACEAE	MALLOW FAMILY							
Sphaeralcea sp.		–	–	–	–	–	*	Sm.

TABLE II—CONT'D

SCIENTIFIC NAME	COMMON NAME	STATIONS 1	2	3	4	5	6	SEASON OF POLLINATION
BORAGINACEAE	BORAGE FAMILY							
Lappula texana	Stickseed	**	–	–	–	–	–	Sm.
VERBENACEAE	VERVAIN FAMILY							
Verbena macdougalii	Vervain	**	–	–	–	–	–	Sm.
LABIATAE	MINT FAMILY							
Pogogyne sp.		–	–	–	–	*	–	Sp. Sm.
COMPOSITAE	SUNFLOWER FAMILY							
Achillea lanulosa		–	–	–	**	–	–	Sm.
Ambrosia psilostachya	Western ragweed	–	***	**	–	**	**	Fl.
Artemisia spiciformis	(identification uncertain)	**	–	**	–	–	***	Fl.
Artemisia camporum		***	–	**	–	–	***	Fl.
Artemisia carruthii		–	–	***	–	***	**	Fl.
Aster subgriseus	Aster	*	–	–	–	–	–	Sm. Fl.
Cirsium sp.	Thistle	–	–	–	–	*	–	Sm.
Coreopsis cardaminefolia	Tickseed	–	–	–	–	–	**	Sm.
Erigeron divergens	Fleabane	**	**	–	–	**	–	Fl.
Franseria tenuifolia	False ragweed	***	–	***	–	–	**	Fl.
Grindelia aphanactis	Tarweed	–	**	**	***	***	**	Fl.
Gutierrezia sarothrae		–	–	–	–	**	–	Fl.
Helianthus annuus	Common sunflower	***	–	–	–	***	***	Sm. Fl.
Hymenoxys sp.		**	–	–	–	–	–	
Lactuca sp.		–	–	–	*	–	–	Sm.
Psilostrophe sparsiflora		–	–	–	–	*	–	Sm.
Ratibida columnifera	Cone flower	–	–	–	–	–	***	Sm.
Solidago glaberrima	Goldenrod	–	**	–	–	–	**	Sm. Fl.
Taraxacum taraxacum	Dandelion	–	–	–	–	*	–	Sm.
Tragopogon porrifolius	Salsify	*	–	–	–	–	–	Sm.

* = present or rare.
** = fairly common, few.
*** = common.
**** = very common or abundant.
– = does not occur.

Fl. = pollinates in the fall.
Sp. = pollinates in the spring.
Sm. = pollinates in the summer.
El. = early in the season indicated.

quite common. Of more significance than these are two of the Chenopodiaceae, *Chenopodium album* and *Salsola pestifer*. *Amaranthus retroflexus*, *Franseria tenuifolia* and *Atriplex rosea* are the only other plants of major importance here.

Station 4, about two blocks southeast of the center of town and the railroad, contains but three important plants: *Muhlenbergia montana*, *Atriplex rosea* and *Grindelia aphanactis*.

Station 5, directly north of town at the edge of the residential section, has a slightly higher elevation than that of the various other

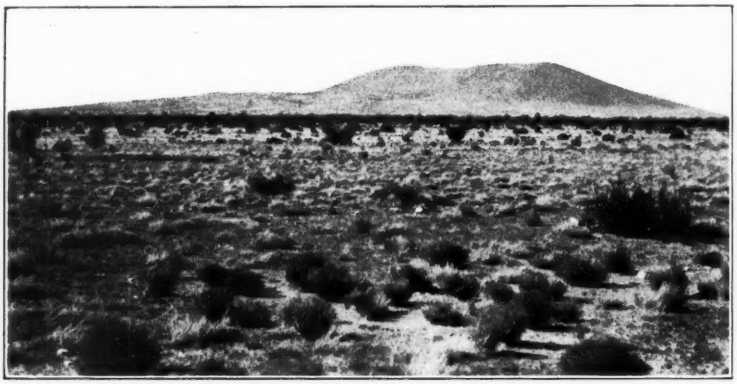

Fig. 5.—The abrupt transition from so-called sagebrush to cedars. The San Franciscan Volcanic Uprising in the background.

Fig. 6.—Pure stand of *Salsola pestifer*, which is limited to the borders of the highway. Some *Artemisia* spp., in the foreground and the cedars in the distance.

sampling points. Here, as in Station 1, *Agropyron tenerum* and *Bouteloua gracilis* are quite abundant. A number of other species are subdominant to them. *Chenopodium album*, *Grindelia aphanactis* and *Helianthus annuus* are fairly common.

Station 6 includes an area beginning just east of the residences and extending along the highway for a distance of six miles in this direction. It has a flora characterized by the *Pinus ponderosa* consociation, which is typical of the entire station except that these have been cleared near the dwellings. Associated with the *Pinus ponderosa* we find *Agro-*

pyron tenerum and *Poa pratensis* as the outstanding Gramineae. Near the town *Salsola pestifer* and *Chenopodium album* are quite abundant, and at the eastern edge of this section *Artemisia* spp. are common.

As one passes eastward from Flagstaff, a relatively abrupt change occurs at a point approximately sixteen miles out. Here the *Pinus ponderosa* becomes much less conspicuous, and *Juniperus monosperma* continues as the dominant for an additional nine miles. Then, even more abrupt is the disappearance of the cedars. This has been referred to in the opening paragraphs. The dark line in the background of Fig. 5 indicates the eastern boundary of this growth. The vegetation in the foreground is devoid of cedars and is characterized by *Artemisia* and *Sporobolus,* excepting a limited area along the highway. This undergoes a gradual transition as one continues eastward toward our sampling Station 8 at Winslow.[3]

As another striking example of the strict limitation of vegetation in this semiarid territory, we wish to call attention to Fig. 6. This was taken from almost the same spot as Fig. 5, but with the camera turned in the opposite direction. The very dense vegetation in the middle of the landscape represents a continuous and nearly impenetrable growth of *Salsola* along the "shoulder" on each side of the transcontinental highway. Rarely is another plant found intruding itself into this growth. On the other hand, a careful search of the area several hundred feet on either side of the highway failed to reveal any *Salsola.* This phenomenon of certain vegetation being sharply limited, in "virgin territory," to that soil disturbed by man has been repeatedly mentioned in several works on botany. Not only is the highway outlined with *Salsola,* but the railroad right of way several miles distant demonstrates the same curious botanic adaptation. This and the previous examples furnish additional evidence to support our often emphasized contention that an accurate botanic survey can seldom be made from an observation of a limited section of the territory involved.

The Williams area is characterized by a dominance, among the trees, of *Pinus ponderosa.* This makes the latter a potential factor in spring pollinosis. Two species of *Bouteloua* and *Aristida fendleriana* apparently pollinate at the same season. The Chenopodiaceae is represented, in the main, by but three species: *Chenopodium album, Salsola pestifer* and *Atriplex rosea.* The last mentioned is somewhat less common than are the previous ones. The Compositae, likewise, has three important species: *Artemisia camporum, Ambrosia psilostachya* and *Gutierrezia sarothrae.* Although the last is perhaps more abundant, it may be of less significance in pollinosis. Other members of the sunflower family occur less commonly.

In Flagstaff, *Pinus ponderosa* similarly occupies a prominent position. The species of Gramineae are less numerous than at Williams. The most

widely distributed are *Agropyron tenerum, Sitanion hystrix, Poa pratensis* and *Lolium perenne.* The last two are especially common in lawns. The identical species of Chenopodiaceae, *Chenopodium album, Salsola pestifer and Atriplex rosea* are prominent here. It should be noted that certain species may be entirely absent from one sampling station and yet practically dominate the next one, but a few thousand yards away. This phenomenon is not limited to members of any particular group or to any single community; although some striking individual examples have been indicated. There is little change from our findings at Williams in the emphasis on the Compositae, except that *Gutierrezia sarothrae* was here noted at but one sampling station, whereas it was the most widely distributed of the Compositae at Williams. More study will be necessary to establish the significance of this plant in clinical allergy.

If one does not have in mind the characteristic botanic findings at Seligman,[2] fifty miles west of Williams, and Winslow,[3] forty-two miles east of Flagstaff, it might be well to review briefly these in comparison with the above observations. These communities, to the west and to the east, are on the lower level of the desert plateau mentioned in the opening paragraphs.

Here, except for the town areas, trees are of little importance; whereas they dominate the montane flora of Williams and Flagstaff. Grasses are of limited distribution and importance in Seligman, but in Winslow the species are much more numerous and several of these challenge other plants for the position of dominant. In Flagstaff the species are still more numerous and of increasing importance, while Williams presents the maximum number of types and perhaps the height of their significance. The forest type of vegetation at the last two places would be expected to include a large variety and a relative abundance of grasses. Very few species are common to the four areas. *Cynodon dactylon,* Bermuda grass, is found in Winslow, wherever there is adequate moisture and other conditions are favorable to its growth. Notwithstanding the greater rainfall in Flagstaff and Williams, careful search has failed to reveal any Bermuda grass in that area; although lawns are not uncommon. We have followed with special interest the variation in occurrence of this grass because it is usually considered the one of major importance in the semiarid regions. It would seem improbable that adequate opportunity had not been afforded to introduce this from Winslow, 42 miles away, or even from more remote areas. In our earliest "scout" surveys we were informed, by one* especially well versed in the botany of Albuquerque, N. M., that Bermuda grass was not to be found in that city. Two years later the same opinion was emphatically repeated and observa-

*Personal communication from Dr. E. F. Castetter, Professor of Biology, University of New Mexico, Albuquerque.

tion of many lawns seemed to support this view. We did, however, observe a small patch of this grass at the railroad station. It seemed to be a pure stand and was apparently introduced as a result of commerce. This patch has been observed for three successive years, and it has now become well established and seems to be spreading. It would appear that we have under observation a botanic invasion, or at least the "advance guard," which may change the entire picture as it relates to pollinosis in that community. How long Williams and Flagstaff will resist a similar invasion is a subject for speculation.

While the variation in trees and grasses is conspicuous, it is overshadowed by that observed among the Chenopodiaceae in the same four communities. Winslow, on the east, is characterized by a large number of species and by the relative abundance of the members of certain of these. We have already described how *Salsola,* limited essentially to four narrow bands—two along the highway and a similar number on the railroad right of way—establishes "liaison" with the Chenopodiaceae at Flagstaff. Interrupted though it is, *Salsola* is the only connecting link between the Chenopodiaceae in the two communities. The three species, representing three genera, which are found in Flagstaff and Williams are of minor significance in Winslow. One of these, *Salsola pestifer,* is the most important of all species at Seligman. It has, therefore, adapted itself to a wide variety of altitude, climate, etc. Apparently the same species is found also in Gallup, N. M.,[4] as well as on the Pacific Coast.

We have previously attempted to indicate the rationale for the use of scientific rather than common names. This does not always suffice to avoid confusion, as we would like to point out in connection with the genus *Salsola.* Our studies have, naturally, had as their basis the botany of California. Jepson,[5] the leading authority, designates the member of this genus as *Salsola kali* var. *tenuifolia* G. F. W. Mey. Hall,[6] another authority on this subject, especially as it relates to pollinosis, uses the term *Salsola kali* without further qualification. He gives its distribution outside of the coastal area as follows: "Apparently very important east of the Sierra Nevada and throughout the Great Basin States." Gray[7] in a more general study is also in accord, as he notes: "It is exceedingly abundant and pernicious in the Northwestern States." Specimens collected by us at Seligman and in other communities to the east are apparently identical with the California variety. In the absence of a survey specifically dealing with north central Arizona we have designated our specimens according to the nomenclature used for the same species in California. In the survey of Gallup, N. M.,[4] we acknowledged the priority of Wooton's[8] work and employed his terminology, *Salsola pestifer.* We are undoubtedly in error in not so designating our specimens in all surveys in Arizona.[2, 3]

Standley[9] may be quoted as the authority who has most recently studied this genus. A few points from his report may be pertinent. Under *Salsola Kali* L. (1753) he lists six synonyms, five of which are considered a variety of *Kali*. *Salsola pestifer* A. Nelson (1909) has at least five synonyms, and three of these include the controversial term *Kali*. One of these, *S. Kali tenuifolia*, may at first glance be assumed to be identical with that described by Jepson; however, Standley gives Tausch and not G. F. W. Mey. as the worker who so designated that specimen. He has not indicated the latter type specimen under either genus. When such authorities on taxonomy are not in accord, we trust that our readers will be somewhat indulgent even though we have employed the two designations for what appears to be a single species.

CONCLUSIONS

1. The San Franciscan volcanic field, including the towns of Williams and Flagstaff, is located at an altitude of 6,500 to 7,000 feet, approximately 1,500 to 2,000 feet higher than adjacent communities on the west and east. Rainfall is likewise in excess of that found at the lower altitudes, even though they are but 40 to 50 miles distant.

2. The vegetation is typically the montane forest type as contrasted with the desert variety surrounding it. *Pinus ponderosa*, *Juniperus monosperma* and *Pseudotsuga mucronata* are the most important trees. Except for the sections where man has disturbed the balance they dominate the landscape. The transition to the desert type is quite abrupt, especially on the east.

3. Grasses are abundantly distributed and numerous species are found. These are not limited to lawns as is sometimes the case in this semiarid region. *Cynodon dactylon* (Bermuda grass) could not be found in this area; although it is rather common in lawns at Winslow, 42 miles east. Albuquerque, N. M., has been cited as another community devoid of this grass, yet it seems to be undergoing an invasion by this plant. We have not explained the evident resistance of Williams and Flagstaff to a similar invasion. This is but another contribution to the insurmountable evidence that the botanic flora of a community is best determined by an actual and rather detailed field study.

4. Members of the Chenopodiaceae are few in number and of limited distribution as compared to the typical desert region. One of these, *Salsola pestifer* maintains a more or less continuous chain of growth from the lower plateau on the west through the higher altitudes of the Williams-Flagstaff area to the basin of the Little Colorado River on the east.

5. The correct botanic designation for Russian thistle, the variety found in California, Arizona, and New Mexico is apparently *Salsola pestifer*. Some of the reasons for the use of other terms have been indicated.

6. The Compositae is represented by but a few species, notably *Artemisia camporum* and *Ambrosia psilostachya*, which seem to be of significance in pollinosis. *Artemisia* spp. extend eastward beyond the boundary of the growth of *Juniperus monosperma*, but become less and less conspicuous as one descends to the altitude of the lower plateau.

7. A comparison of the botany of the area represented by this study, with that on the west and on the east, reveals little similarity. A few outstanding examples are cited above.

In this and the previous survey the herbarium of the Department of Botany at the University of Southern California and Dr. Freda Detmers, its curator, have contributed much in the identification of specimens. Also, great credit is due Mr. D. L. Smith, superintendent of the Santa Fe Hospital, for his timely advice and assistance.

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STUDIES IN HAY FEVER

Clinical Observations Including a Botanical and Air Survey of the Utah Region

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WITHIN recent years the environmental factor in the study of hay fever has received increasing attention among medical men. It has been shown that for effective treatment specific desensitization against the pollen encountered is essential and that desensitization against a pollen not encountered by the patient is of little avail to him no matter how highly sensitive to it he may be. As a consequence the study of the flora adjacent to large centers has become necessary in order to obtain information leading to more successful treatment.

It is now generally accepted also that the only pollens which are significant factors in the etiology of hay fever are those which are wind borne in contradistinction to those which are insect pollinated.

Coca, Walzer and Thommen[1] in their able treatise on hay fever lay down five postulates which a pollen must fulfill before it can be considered a cause of hay fever:

(1) It must contain an excitant for hay fever. (2) It must be anemophilous, or wind borne, as regards its mode of pollination. (3) It must be produced in sufficiently large quantities. (4) It must be sufficiently buoyant to be carried considerable distances. (5) It must be produced on an abundant, widely distributed plant.

With these postulates in mind, it is more important than ever that careful and reliable botanical surveys be made before allergists can successfully relieve hay fever in any given district. The last of Coca, Walzer, and Thommen's conditions is of the greatest significance to the practicing physician because the pollens, regardless of their physical characteristics, are, of course, of no significance if the plants bearing them are not widely and abundantly distributed in the patient's environment. Thus, in the West the sages are of the utmost importance, while in New England they are of no practical significance, although many varieties have been reported by House[2] in New York state. Russian thistle, one of the greatest single causes of hay fever in the intermountain states, the writer has gathered and identified within the campus of the University of Chicago. It has also been found, together with other such western prime offenders as the amaranths, atriplexes, and chenopods, within a few miles of New York City by Taylor.[3]

In Chicago and New York these plants are of no importance in the etiology of hay fever because of their very limited distribution.

That the hay fever problem varies widely in different parts of the country, as does the flora, is evident from the literature. Black[4] in Texas reports a high percentage of relief in forty patients treated with cedar pollen extracts. Henry and Herring[5] surveyed Memphis and its environs and found the ragweeds the chief cause of hay fever, while Watry and Lamson[6] reported no ragweed present in Needles, California, and credited the hay fever there primarily to Bermuda grass with the atriplex, or salt bush group, close seconds and the willows and cottonwoods minor offenders. French[7] found the pollens of south central Texas divided into four groups as regards their responsibility for hay fever: grasses cause from 35 per cent to 40 per cent; ragweeds from 35 per cent to 44 per cent; careless weeds from 16 per cent to 23 per cent; and plantain and mountain cedar, 5 per cent. Piness and Miller[8] studied 133 patients in a mining community and found them sensitive to the various pollens as follows: Bermuda grass, 80 per cent; Russian thistle, 79 per cent; careless weeds, 65 per cent; atriplexes, 56 per cent; salt grass, 35 per cent; and Johnson grass, 27 per cent. In New Orleans, according to Penfound, Efron and Morrison,[9] the various groups of hay fever producing plants rank in relative importance as follows: grasses, 100; ragweeds, 29; composites, 10; amaranths, 5; chenopods, 4; and docks, 2.

Detweiter and Hurst[10] believe that treatment should be directed toward the elm, oak and birch in the spring, the grasses in the summer, and the ragweeds in the autumn.

On the basis of atmospheric pollen entrapped on glass slides in a study made in Chicago, Koessler and Durham[11] distribute the plants as follows on a basis of effective pollen dissemination. (Trees are not included.)

Ragweeds	65.3%
Grasses	21.4%
Chenopods	4.2%
Amaranths	2.1%
Docks and plantain	2.0%
Composites	1.0%
Hop and hemp	1.0%
All others	3.0%

Thommen himself in his discussion of the hay fever situation in New York lists the following as of primary importance: Trees: oak, hickory, birch, ash and beech. Grasses: sweet vernal, June, orchard, red top, and timothy. Weeds: plantains and ragweeds.

For purposes of study Stier, Hollister and Bonser[12] divided the area of the Pacific Northwest into seven districts and studied the plants according to the geological formations in the districts.

The following botanical surveys are of note: Berton's[13] Study of the Anemophilous Plants of the District of Columbia; Durham's[14] Sur-

vey of Indiana; Piness, Miller and McMinn's[15] Report on the Botany of Southern California; Balyeat's[16] Survey of Oklahoma; Kahn's[17] Botany of Southwest Texas; Phillip's[18] Study of Central Arizona; Rowe's[19] Report on the San Francisco Bay Region; Hall's Hay Fever Plants of California[20]; Watson and Kibler's[21] Study in Arizona and the Southwest. Others have also been made by Templeton[22] and Piness[23] in California; Mullin,[24] Waring,[25] Conway,[26] and Wolf[27] in Colorado; Foss[28] in Montana; Chamberlain[29] and Faskett[30] in Oregon; Spitz[31] and Figley[32] and Rackeman[33] in New England. Anderson[34] gives a list of hay fever plants encountered in Utah but gives no data as to their abundance.

Of the work done in Europe, the outstanding contributions are those of Blackley[35, 36] in England; Freeman,[37] Hansen,[38] and Loeb and Petow[39] in Germany; Benjamins, Idzerda and Uittien[40] in Holland; and Boagoe[41] in Denmark.

AIR ANALYSIS

Descriptions of pollinosis have come down to us from the Middle Ages. The first reference in medical literature is made by Botallus[42] in 1565 when he refers to the odor of roses as a cause of severe distress. Roses were also blamed for summer catarrh by Ledelii[43] in 1683, Riedlini[44] in 1695, and others. The term hay fever made its first appearance in medical literature in 1828 when Bostock[45] used it. But it remained for Blackley[35, 36] in 1873 to clarify the subject. Indeed so painstaking were his scientific studies and investigations covering a period of fourteen years, that his book on hay fever is one of the outstanding pieces of research not only for his day but for all time. In fact this first book of his,[36] together with his second in 1880,[35] leaves little for the research worker of today to add.

Blackley's work was so far ahead of his time that no effort was made even to prove his researches until Dunbar,[46] a physician of Hamburg, published his monograph on hay fever in 1903.

Since Dunbar's contributions, literally hundreds of workers have applied themselves to the cure and prevention of hay fever and Blackley's experiments have been repeatedly verified.

In order that the various workers might have a uniform yardstick to use in comparing their experimental evidence, Scheppegrell[47] in 1917 devised a formula for estimating the pollen content of the air in grains per cubic yard by counting the amount falling on one square centimeter of coated glass slide per twenty-four hours. In this formula

$$n = \frac{7 \times N}{v \times t}$$

when n is the pollen content of the air in grains per cubic yard; N, the number of pollen grains falling on each square centimeter of

glass slide; v, the velocity of fall for the pollen under consideration; and t, the number of hours of exposure. The values of v were determined for pollen of different diameters and are presented by Scheppegrell. Since devising this formula he has made other noteworthy studies on atmospheric pollen.[48, 49, 50]

By the use of Scheppegrell's "yardstick," Durham[51] with the cooperation of over 30 physicians in representative cities in the United States has reported values for ragweed pollen in the atmosphere of the following cities in 1927.

	MAXIMUM CONCENTRATION POLLEN GRAINS PER CUBIC YARD	DAILY AVERAGE
Oklahoma City	3,600	814
Indianapolis	2,100	470
Kansas City	2,237	400
Chicago	250	100
New Orleans	410	75
New York	100	26
Philadelphia	38	10
Richmond, Virginia	12	6
Oakland, California	6	1

In 1927, Thommen[1] made a study of the ragweed concentration of the atmosphere over New York City and found a maximum count of 105 grains per cubic yard of air. He also tabulated the precipitation, wind velocity, maximum temperature, sunshine and humidity, and found that the factors most influencing the amount of pollen in the air are sunshine, wind velocity and precipitation.

Because of the fact that ragweed pollen is the chief cause of hay fever, practically all the reports thus far published have stressed the ragweed pollen content of the air.

Through the assistance of the United States Weather Bureau and the Canadian Meterological Service, Durham[52] recently had slides exposed in thirty United States and four Canadian stations. All of these slides were sent to a central office and the entrapped ragweed pollen was counted. The result showed that the ragweed pollen concentration is very high in the Middle West and gradually becomes less in the eastern states. In the West and on the Pacific Coast it is relatively scarce. Seasons with abundant rainfall favored the production of ragweed pollen as well as other more useful crops.

Koessler and Durham[11] made an air analysis of Chicago which demonstrated that the grass and ragweed pollens accounted for nearly 87 per cent of the total atmospheric contamination. This is essentially in agreement with the conclusions of Duke and Durham.[53] Many other observations of value were made by Durham.[54, 55, 56]

Detweiler and Hurst[10] in their study of atmospheric pollen found elm, oak and birch pollen predominant in the early spring, the grasses in the summer, and ragweed in the autumn.

The first report of pollen counts made throughout the entire twelve months of the year was made by Rowe[19] for the San Francisco Bay region. He, as others, found the grass pollens most abundant. According to his results, there is no period in San Francisco when atmospheric pollen is entirely suppressed except on days of constant rain.

THE SALT LAKE CITY STUDIES

No reports were available as to the wind-borne pollens of the Rocky Mountain region, and so a botanical survey of the region surrounding Salt Lake City was commenced in 1928 and pushed vigorously during the summers of 1929 and 1930. Six test plots were chosen near Salt Lake City. These plots were visited twice weekly and a record was kept of all the plants found, as well as the dates of pollination and the relative amount of pollen shed. A detailed list of the results is shown in Table I. A very fair representation of the flora of the entire intermountain states was obtained since these plots, as indicated below, contained alkali flat, desert, swamp, canyon, farming land, and high grade residence property.

Plot I, the City Creek Plot, is situated at the mouth of City Creek Canyon. It is about one mile long by one-quarter mile wide. About half of it is bottom land with a stream flowing through it, the other half is hillside gradually rising to an average height of 700 feet above the creek bed. On account of the light rainfall in this region the hillside is almost desert in character and bears several varieties of cactus. The business district is about one and one-half miles from this plot.

Plot II, the North Bench Plot, comprises a more or less vacant area on the north bench extending from H Street to the cemetery and north to the foothills. It is on the outer edge of the residence district and faces south.

Plot III, the Hot Springs Plot, about two miles from the business district, is about a mile square and extends from Beck's Hot Springs north along the highway and west in the alkaline flat for a mile. One-half of the plot, the soil unsuitable for cultivation, carries a typical alkaline flora; the other half is swamp land.

Plot IV, the Jordan River Plot, about a square mile in area, reaches from Second South to Fourth South along the Jordan River and west to the airdrome. It is partially under cultivation and partially virgin territory overgrown with alkaline vegetation. This plot is about one and one-half miles west of the business district.

Plot V, the Gilmer Park Plot, comprises about a half square mile in the center of Salt Lake City's finest residential district, Gilmer

TABLE I

BOTANICAL SURVEY OF SALT LAKE CITY

PLANTS	DATE OF POLLINATION		ABUNDANCE IN PLOT: 1	2	3	4	5	6
Acer negundo	April	Plant:	++	+	+	+	+	+
(Box elder)		Poll.:	++++	++++	++++	++++	++++	++++
Acer grandidentatum	April	Plant:	++			+		
(Mountain maple)		Poll.:	++			++		
Populus alba	April	Plant:	++		++		++	
(White poplar)		Poll.:	+		+		+	
Populus deltoides	April	Plant:	++	+	+			
(Lombardy poplar)		Poll.:	++	++	++			
Ulmus americana	April	Plant:		+	+			++
(American elm)		Poll.:		+	+			++
Quercus gambellii	May	Plant:	++++	+				
(Scrub oak)		Poll.:	++++	++++				
Carex aquatica	April–May	Plant:			+	+		+++
(Sedge)		Poll.:			+	+		+
Bromus tectorum	May–June	Plant:	++++	++++	++++			
(June grass)		Poll.:	++	+	+			
Dactylis glomerata	May–June	Plant:		+		+	++	++++
(Orchard grass)		Poll.:		+++		+++	+++	+++
Poa pratensis	May–June	Plant:	++	++++	++	+++	+++	+++
(Blue grass)		Poll.:	++	+++	++	++	++++	++
Scerpus palidosus	May–June	Plant:			++++	+++		
(Bullrush)		Poll.:			++	++		
Rumex occidentalis	May–June	Plant:		+	+	+	+	+
(Western dock)		Poll.:		++	++	++	++	++
Festuca elatior	May–June	Plant:				++		+
(Fescue)		Poll.:				++		++
Phleum pratense	June–July	Plant:	++			+	+	+
(Timothy)		Poll.:	++			++	++	++
Catabrosa aquatica	June–July	Plant:			++++			
(Mountain grass)		Poll.:			++			

TABLE I—CONT'D

PLANTS	DATE OF POLLINATION		ABUNDANCE IN PLOT: 1	2	3	4	5	6
Agropyrum subvillosum	June–July	Plant:	++		++	+++		++
(Couch grass)		Poll.:	++		+	+		+
Distichlis spicata	June–July	Plant:			++++	++++		
(Salt grass)		Poll.:			++	++		
Iva axillaris	June–July	Plant:			++	++		
(Poverty weed)		Poll.:			++	++++		
Hordeum jubatum	June–July	Plant:			++	+++	++	+
(Fox tail)		Poll.:			+	+	+	+
Atriplex truncata	June–July	Plant:			++++	++++		
(Annual saltbush)		Poll.:			+++	++		
Plantago lanceolatus	June–July	Plant:						++
(English plantain)		Poll.:						++
Medicago sativa	June–August	Plant:		+	+++		+++	
(Alfalfa)		Poll.:		+	++		+	
Atriplex canescens	June–August	Plant:			++			
(Shadscale)		Poll.:			+			
Atriplex hastata	June–August	Plant:			++	++		
(Shadscale)		Poll.:			+	++		
Melilotus alba	July–August	Plant:	++	+++	+	+	++++	+++
(White sweet clover)		Poll.:	+	+	+	+	+	+
Salsola pestifer	July–August	Plant:	++++	++++	+++	++	+++	++++
(Russian thistle)		Poll.:	+++	+++	++	+++	+++	++++
Chenopodium album	July–August	Plant:			+	++	++	
(Lambs quarter)		Poll.:			+	++	++	
Chenopodium glaucum	July–August	Plant:			+++	+++	++	
(Lambs quarter)		Poll.:			+	++	++	
Kochia scoparia	July–August	Plant:				++		
(Summer cypress)		Poll.:				++		
Xanthium echinatum	July–August	Plant:			+++		++++	++
(Cocklebur)		Poll.:			+		+	+

TABLE I—CONT'D

PLANTS	DATE OF POLLINATION		ABUNDANCE IN PLOT: 1	2	3	4	5	6
Grindelia squarrosa	July–September	Plant:				+	+++	+++
(Gum plant)		Poll.:				+	+	+
Chrysothamnus graveolens	August–September	Plant:	+++					
(Rabbit brush)		Poll.:	+++					
Guitterizia microfolia	August–September	Plant:	+++					
(Rabbit brush)		Poll.:	+++					
Artemisia wrightii	August–September	Plant:	++	+			++	+
(Prairie sage)		Poll.:	++	++			++	+
Artemisia tridentata	August–September	Plant:	++++	++++	++	+++	+	++
(Black sage)		Poll.:	++++	++++	++	++	++	++
Ambrosia elatior	August–October	Plant:	++	++	+++	++	+	+
(Common ragweed)		Poll.:	++	++	+++	++	+	+
Iva xanthifolia	August–October	Plant:			+++	+		
(Burweed marsh elder)		Poll.:			+++	+++		
Franseria acanthacarpa	August–October	Plant:			++++			
(False ragweed)		Poll.:			+++			

Park, and is about one and one-half miles from the business district.

Plot VI, the Country Club Plot, contains about 40 acres, including a golf course, residence property and a considerable amount of truck-gardening land. The area is irrigated and growth is luxuriant, both of crops and of grasses and weeds along the roadways and the margins of the plot.

These studies showed that ragweed is neither the most abundant plant in the intermountain region nor the heaviest contaminant of the air; nor does it appear as the chief cause of hay fever as determined by cutaneous tests on hay fever patients. As illustrated in Table III, the four pollens of primary importance in this region are the Russian thistle, the sages, the grasses and the ragweed, respectively. Consequently a total atmospheric pollen survey of Utah as a typical intermountain state was undertaken, rather than merely a ragweed survey. Pollen-counting stations were established at Salt Lake City, Ogden, Logan and Provo, the greatest distance between them being 150 miles.

Salt Lake City is situated very close to the Wasatch Mountains on the east with the greater part of the city within two miles of the mountains or high foothills. To the north and south are found extensive areas of irrigated land largely devoted to fruit and truck gardening. For nearly fifteen miles to the west a virgin alkaline flat is covered with semidesert and alkaline-loving plants.

Logan, Ogden and Provo are agricultural communities surrounded by highly cultivated, irrigated land except on the east where the mountains and foothills rise several thousand feet and are covered with sagebrush and, at some seasons, with grasses and desert weeds.

Pollen-counting stations were established in each town. In Salt Lake City and Ogden the stations were on the roofs of office buildings; in Logan and Provo they were in the more residential sections. The Budge Memorial Hospital in Logan and Dr. Merrill and Dr. Oaks in Provo cooperated in this survey by exposing the slides.

The following uniform technic was used in counting the pollen entrapped on the slides. With a 10 ocular and a 16 mm. objective and draw tube length of 160 mm., there is an area of the visual field of 1.51+ sq. mm. Counting 66 separate fields in four or five parallel sweeps over an ordinary one-inch by four-inch glass slide gives an area of 99.66 sq. mm. which is near enough to a square centimeter for purposes of this survey and is well within the error of observation. All slides were counted by the same technician.

The computations as to the content of the air in grains per cubic yard were made by use of the formula of Scheppegrell already referred to. In these computations the grasses were considered to have a uniform diameter of 35 microns (red top, 30; blue grass, 33; orchard

grass, 36; and timothy, 40); Russian thistle, 25; sagebrush, 25; and common ragweed, 18. Taking into account the relative pollen abundance as shown in Table II and the total length of the pollinating period as determined by this survey, the following relative values were used for purposes of determining an average pollen grain size: grasses, 10; Russian thistle, 8; sage, 7; and ragweed, 6. By multiplying the pollen sizes by these figures and taking the average, 23.2 microns is determined as the size of an average pollen grain for this region. This is 23 per cent smaller than the smaller grass pollens and 29 per cent larger than ragweed pollen, and represents a fair average size for the pollen grains in the air of the intermountain states.

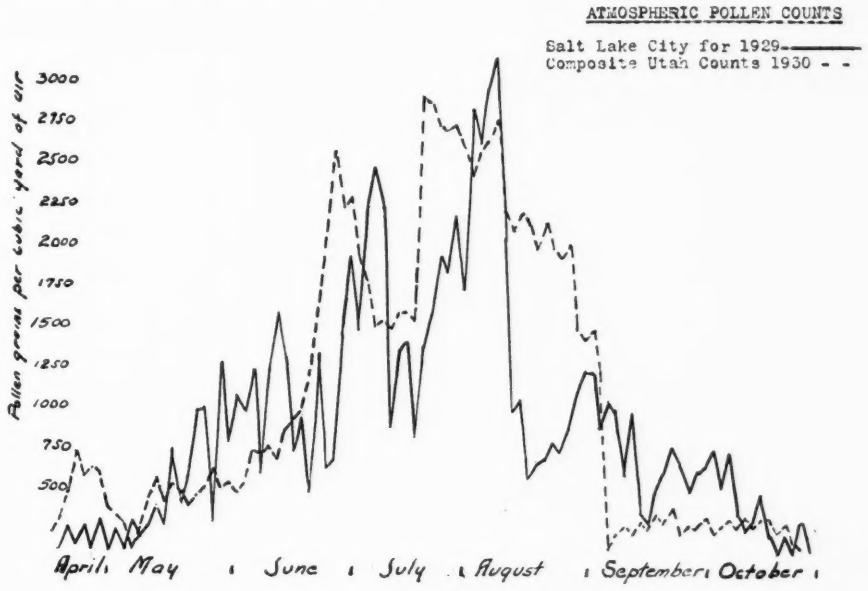

Chart 1.

For this study, the usual one inch by four inch microscope slides were coated with a very thin layer of petroleum jelly and exposed in a horizontal position for twenty-four hours. The place of exposure was in all cases at least fifteen feet from any obstruction. Because of the infrequence of summer rains in this region no precautions were taken to prevent damage from that source. The counts per square centimeter and an average pollen grain size of 23.2 microns, determined as explained above, were used in Scheppegrell's formula and the total pollen content of the air was computed.

RESULTS

Plotting the counts of the four towns, a general similarity was noted, the only differences being in the height of the curve and the dates of their peaks. Chart 1 shows graphically the curve for Salt

Lake City for 1929 and the composite curve of the four towns for 1930. Both peaks fall in late July and early August, at which time the grasses are beginning to wane and the weeds to reach their maximum. Secondary peaks occur in September, due to the pollination of the thousands of acres of sages (artimesias) which cover the West. The ragweeds pollinate from early August until the first killing frost in October.

The short sharp rise in April, 1930, is due to the abundance of maple and box elder pollen, which is very heavy when the weather is warm and dry. Nearly half the shade trees lining the streets of Utah towns are maple or box elders.

An analysis of these graphs reveals a peak count of 3,078 grains per cubic yard which is several hundred less than the peak Durham reported for Oklahoma City for ragweed alone (3,600). It is, however, much higher than Durham's peak figure for ragweed alone in Salt Lake City.

At the close of the season, the "field notes" were tabulated to determine plant abundance and pollen production. No attempt was made at first to get figures on atmospheric pollen distribution—only an estimate of the amount of pollen produced, whether liberated into the air or merely settling to the ground within a few feet of the parent plant. By counting the number of plants in a definite proportion of the area studied, an estimate was made of the total plant population. A record was made also of the length of the pollination period. During the summer one or two plants of each variety were cut off at the roots, the cut stems kept wet with rags, and the total pollen shed in twenty-four hours was weighed. With the total plant population, the length of the pollinating period, and the total amount of pollen shed in twenty-four hours estimated, an estimate of the total pollen production became possible and was made.

In order to have a basis for comparison, the plant and pollen production of the common ragweed was given an arbitrary value of 100 and the other plants were rated on this base. By adding the plant and the pollen production values together, a third arbitrary value called the composite rating was obtained and the plants were listed accordingly, as shown in Table II.

From Table II it will be seen that the ragweeds are not the important hay fever producing plants in this region which they are in the central and eastern states. Grasses, sages, and Russian thistle outrank them. From the standpoint of pollen production and plant distribution, the most important anemophilous plants of the intermountain states rank as follows: (1) *Graminaceae grasses.* (2) *Chenopodaceae*—Russian thistle, saltbushes, summer cypress. (3) *Artemisia*—sages. (4) *Ambrosiaceae*—ragweeds.

As in Penfound, Efron and Morrison's survey of New Orleans, and Koessler and Durham's work in Chicago, the grass group leads the list. In both these reports the ragweeds are second as contrasted with

TABLE II

RELATIVE ABUNDANCE AND POLLEN PRODUCTION OF THE PLANTS OF GREATEST IMPORTANCE*

NO.	COMMON NAME	SCIENTIFIC NAME	RATING		
			PLANT	POLLEN	COMPOSITE
1	Blue (June) grass	*Poa pratensis*	150	175	325
2	Red top	*Agrostis alba*	160	150	310
3	Russian thistle	*Salsola pestifer*	210	95	305
4	Sagebrush	*Artemisia tridentata*	180	85	265
5	Orchard grass	*Dactylis glomerata*	115	145	260
6	Common ragweed	*Ambrosia elatior*	100	100	200
7	Quack grass	*Bromus tectorum*	160	10	170
8	Annual saltbush	*Atriplex truncata*	80	75	155
9	Couch grass	*Agropyrum pseudorepens*	110	40	150
10	Oak	*Quercus gambellii*	60	90	150
11	False ragweed	*Franseria acanthacarpa*	80	65	145
12	Summer cypress	*Kochia scoparia*	75	65	140
13	Cottonwood	*Populus angustifolia*	50	90	140
14	Shadscale	*Atriplex canescens*	75	60	135
15	Burweed marsh elder	*Iva xanthifolia*	40	60	100
16	Alfalfa	*Medicago sativa*	95	1	96
17	Rabbit brush	*Chrysothamnus graveolens*	85	10	95
18	Saltgrass	*Distichlis spicata*	75	20	95
19	Fox tail	*Hordeum jubatum*	80	2	82
20	Balsam root	*Balsamorhiza sagittata*	80	1	81
21	Samphire	*Allanrolphia occidentalis*	65	15	80
22	Timothy	*Phleum pratense*	20	60	80
23	Dandelion	*Taraxacum officinale*	65	10	75
24	Sueda	*Sueda spp*	60	5	65
25	Box elder	*Acer negundo*	20	45	65
26	Poverty weed	*Iva axillaris*	50	10	60
27	Sedges	*Carex spp*	45	5	50
28	White clover	*Melilotus alba*	35	5	40
29	Lambs quarter	*Chenopodium glaucum*	30	10	40
30	Pigweed	*Amaranthus retroflexus*	25	5	30
31	Prairie sage	*Artemisia ludoviciana*	20	10	30
32	Rattlesnake grass	*Bromus marginatum*	15	1	16
33	Sunflower	*Helianthus annuus*	15	1	16
34	Elm	*Ulmus americana*	10	1	11
35	Pepper grass	*Lepidium perfoliatum*	10	1	11
36	Beard grass	*Polypogon monspeliensis*	10	1	11
37	Fescue	*Festuca elatior*	10	1	11
38	Cocklebur	*Xanthium echinatum*	5	2	7
39	Dock	*Rumex spp*	5	1	6
40	Manna grass	*Glyceria nervata*	5	1	6
41	Plantain	*Plantago major*	3	1	4
42	Ash	*Fraxmus americana*	3	1	4
43	Ragwort	*Lenicio perplexus*	1	1	2
44	Cat-tail	*Typha latifolia*	1	1	2
45	Greasewood	*Sarcobatus vermiculatus*	1	1	2

*The abundance and pollen produced by the common ragweed is given an arbitrary value of 100. Composite rating is the sum of the other ratings. Ratings of one or less are not listed.

fourth in the West where chenopods and the wormwoods outrank them; the wormwoods have a very limited distribution east of the Mississippi.

Russian thistle, the most prolific plant in the Salt Lake City section, ranks fifth in pollen production but its extremely high toxicity accounts for the fact that over 54 per cent of the hay fever patients are sensitive to it and that over 74 per cent of those suffering from late hay fever are Russian thistle sensitive as shown in Table III.

TABLE III

PATIENTS REACTING TO VARIOUS POLLENS

NO.	COMMON NAME	SCIENTIFIC NAME	NUMBER	PERCENTAGE
1	Russian thistle	*Salsola pestifer*	980	54.4
2	Sagebrush	*Artemisia tridentata*	613	33.7
3	Common ragweed	*Ambrosia elatior*	576	31.4
4	Prairie sage	*Artemisia ludoviciana*	487	27.0
5	Timothy	*Phleum pratense*	476	26.4
6	Summer cypress	*Kochia scoparia*	465	25.8
7	Orchard grass	*Dactylis glomerata*	463	25.6
8	Burweed marsh elder	*Iva xanthifolia*	455	25.2
9	Blue (June) grass	*Poa pratensis*	434	24.1
10	Box elder	*Acer negundo*	382	21.1
11	Lambs quarter	*Chenopodium glaucum*	362	20.1
12	False ragweed	*Franseria acanthacarpa*	346	19.1
13	Maple	*Acer spp*	345	19.1
14	Annual saltbush	*Atriplex truncata*	293	16.3
15	Red top	*Agrostis alba*	252	14.0
16	Poverty weed	*Iva axillaris*	251	13.8
17	Pigweed redroot	*Amaranthus retroflexus*	224	12.6
18	Cottonwood	*Populus angustifolia*	129	7.2
19	Shadscale	*Atriplex canescens*	111	6.2
20	Saltgrass	*Distichlis spicata*	105	5.3
21	Rabbit brush	*Chrysothamnus graveolens*	96	5.3
22	Samphire	*Allanrolfia occidentalis*	87	4.8
23	Dandelion	*Taraxacum officinale*	72	4.0
24	Winter fat	*Eurotia lanata*	38	2.1
25	Elm	*Ulmus americana*	26	1.6

The tree hay fever season is always short and is but a minor factor in most places. This is true here of the scrub oak which is extremely abundant on the hills and mountain sides and which produces pollen luxuriantly; indeed it ranks sixth in pollen production. Its toxicity is, however, very low, for results proved that but 0.4 per cent of patients reacted to it. On the other hand, the box elder which ranks much lower in abundance and pollen production was found to account for 21.1 per cent of the patients suffering from hay fever. This is partially explained by the fact that the box elder is the West's principal shade tree and its pollen must fall a considerable distance before reaching the ground, allowing for much greater dissemination and for its entering the patients' sleeping quarters in rather large amounts and exposing the patients for long periods.

It has been the practice to desensitize patients suffering from grass hay fever with timothy pollen extract because this pollen is usually accepted as the most important of the grass pollens. According to Table II, timothy ranks twenty-second in the intermountain states

whereas blue grass, red top and orchard grass are among the first five. From Table III, however, it appears that there are more patients sensitive to timothy than to any of the other grasses. The proportion is as follows: timothy, 26.4 per cent; blue grass, 24.1 per cent; and orchard grass, 25.6 per cent. Of interest in this connection is the fact that although the western hills are literally covered with quack grass in the early spring, its pollen production and toxicity factor are so low that but 0.2 per cent of the hay fever population are sensitive to it.

Here, as everywhere, the ragweed pollen must be taken into account because of its toxic effect. This is convincingly shown by the fact that although it ranks tenth in distribution and fifth in pollen production, it is third highest in the number of patients credited as reacting to it. Apparently the distribution is the only factor which prevents the ragweed from assuming as important a position in this region as it does in the eastern states.

Another interesting factor in sensitivity is found in the study of the two important sages encountered in the intermountain section. The prairie sage has but one-ninth the distribution, produces one-eighth of the pollen, and nevertheless has four-fifths as many patients to its credit as its ubiquitous rival, the sagebrush.

The sedges, although moderately abundant, are of little importance, for less than 0.1 per cent of the patients are sensitive to them.

For a few weeks in the early spring dandelions actually make the fields and lawns of some sections of the West a golden carpet. The plant is insect pollinated but its pollen is highly toxic as revealed by the fact that 4 per cent of the hay fever sufferers are sensitive to it. Seven cases have been encountered in which it was the sole or major factor, and complete relief was obtained by specific desensitization.

Pollen was collected from all the plants listed in Table I, and extracts were prepared. All pollen was collected from the intact plants cut in the late afternoon and placed in a horizontal position over glazed paper, the cut ends of the plant stems being kept moist with wet rags. After from thirty-six to forty-eight hours all the pollen that could be dislodged from the flowering heads by gentle beating was collected and cleaned by sifting. The pollen used was passed through a sieve of 200 meshes to the inch and then defatted with ether. Two per cent solutions were made in a buffered glycerosaline solution and passed through a Seitz filter. Enough phenol to make a 0.5 per cent solution was added. More than 1,800 patients were tested with these extracts by means of the scratch test. Although the intradermal test was used occasionally for experimental purposes, it was not used on any of the cases reported in this series. All reactions smaller than that obtained by the scratch test from a histamine solution of 1:5,000 were considered as one plus or less.

The experience gained from these tests, using those with the histamine solution as positive controls, leads to the belief that they are reliable tests as to the patient's skin sensitivity. Almost invariably patients giving large histamine wheals gave much larger wheals from the pollen extracts than were average. When about 300 patients had been tested, it was found that a large number of the pollen extracts were causing very few, if any, reactions. At this time, all those pollens to which less than 1 per cent of the patients were found sensitive, were discarded and the tests were continued only on those listed in Table III.

TABLE IV

POLLEN SENSITIVITY

PATIENTS' SENSITIVITY	NUMBER	PERCENTAGE
Sensitive to one pollen	174	9.7
Sensitive to two pollens	278	15.4
Sensitive to three pollens	462	25.7
Sensitive to four or more pollens	887	49.2
	1,801	100.0
Sensitive to tree pollens	332	33.1
Sensitive to grass pollens	746	41.4
Sensitive to weed pollens	1,263	70.1
Sensitive to trees only	127	7.0
Sensitive to grasses only	308	17.1
Sensitive to weeds only	789	43.8
Sensitive to more than one group	577	32.1
	1,801	100.0

These same reaction charts were grouped according to patients instead of pollens, as shown in Table IV. One of the striking results was the fact that there were very few individuals whose hay fever was due to a single pollen.

Although 33 per cent of the patients were tree pollen sensitive, only 7 per cent were sensitive to tree pollen alone. Forty-one per cent were grass sensitive, although hay fever was due solely to grass pollens in only 17 per cent of the group. The multiple sensitivity of hay fever patients is well portrayed in this table. The logical assumption from the results obtained in these studies is that effective treatment must be directed to *multiple* desensitization, not merely desensitization.

DOES TRAVEL INFLUENCE HAY FEVER SYMPTOMS?

While the Salt Lake City studies were being made, an incidental study was made of the effect of travel on hay fever symptoms. By close observation of a large number of railway trainmen it was conjectured that they suffered from hay fever much more severely when on duty than when at home. The cooperation of several trainmen patients was enlisted in order to obtain accurate data on the subject.

These men exposed coated slides on steam trains entering and leaving Salt Lake City. The slides were distributed on the locomotive, in the chair cars, in the Pullman cars, and on the observation platform. I made similar exposures in the Bamberger Electric Railway and in my private automobile.

The exposures were for varying lengths of time and a total pollen count per cubic centimeter was determined on each side. As a basis for comparison, the pollen count made the same day in Salt Lake City was used. The counts made on the trains and in the automobile were equated on a twenty-four-hour basis by multiplying the actual count by a figure representing the fractional part of the day during which the exposures were made. During both the day and the night slides were exposed on the trains. The same man who placed slides in the cars at designated points on the trip out of Salt Lake City, collected them one or two hours later at other designated points. Slides were then exposed between the same stations on the return trip, usually twenty-four to thirty-six hours later. I made daily trips between Salt Lake City and Ogden, an hour's ride on the electric train or by auto, and exposed slides in the same manner as the trainmen did. The results are shown in Table V. During the coming summer additional counts are planned in an attempt to correlate the speed of the car with the amount of pollen entrapped. This may be accomplished by arranging for two cars to leave the office at the same time to cover the same route, one at an average speed of 40 to 45 miles an hour and the other at 25 to 30 miles an hour.

With the pollen counts in Salt Lake City as a basis for comparison, it was determined that a hay fever patient would inhale each hour 2.7 times as much pollen in a Pullman coach as in town, 3.8 times as much in the chair car, and over eight times as much on the observation platform. Granting for argument that the development of symptoms in the hay fever patient is in direct proportion to the amount of pollen inhaled, it was found that in driving in an automobile with all the windows open he would be 4.7 times as likely to suffer as he would be at home, while in the electric train his chances would be 6.2 times as great. Should he be an engineer or a fireman on a locomotive, his tolerance would have to be 7.2 times as high to avoid symptoms as it would have to be at home.

The table shows July 6 as the date of highest pollen concentration. On this day there was found to be nine times the pollen concentration of the city on the locomotive slide, nearly ten times on the observation car slide and five times on the coach. The Pullman slide was broken. The counts on the electric train were, however, average; this may in part be explained by the fact that the counts on the electric train were made north of the city and those on the steam road, south.

TABLE V

POLLEN COUNTS MADE FROM COATED SLIDES EXPOSED ON VEHICLES OF VARIOUS KINDS

Slides exposed in the cab of the locomotive, window ledge of coach, Pullman and electric interurban cars, platform of the open observation car, and arm rest of the back seat of the sedan with all windows open. Pullman screens in place, coach and interurban, usually some windows open.

DATE	PLACE OF EXPOSURE (COUNTS EQUATED TO 24 HOURS' EXPOSURE)												COUNTS SALT LAKE CITY
	LOCOMOTIVE		DAY COACH		PULLMAN		OBSERVATION		ELECTRIC INTERURBAN		AUTO (SEDAN)		
	HOURS	COUNT	HOURS	COUNT	HOURS	COUNT	HOURS	COUNT	HOURS	COUNT	HOURS	COUNT	
June													
16	2	8,128	2	4,960		-----		-----	1	12,672		-----	1,074
17		-----		-----		-----		-----		-----	1	6,331	1,310
20		-----		-----		-----		-----	1	15,840		-----	1,110
22	2	6,594	2	4,168	2	4,704		-----	1	14,618		-----	1,680
23	1	7,844	1	4,752	1	-----	1½	11,088		-----	1	6,902	1,778
25		-----		-----		-----		-----	1	9,524		-----	1,662
27		-----	1	9,206	1	4,638		15,784		-----	1	8,740	2,072
July													
6	1½	19,208	1½	11,880		-----	1½	20,671	1	8,478		-----	2,138
7		-----		-----		-----		-----		-----	1	4,786	1,542
9		-----		-----		-----		-----	1	7,814		-----	1,356
10		-----		-----		-----		-----	1	10,876		-----	1,680
14	2	5,336	2	4,238	2	3,876	2	7,928		-----	1	6,330	1,722
15	2	11,082	2	4,708	2	3,584	2	6,134		-----		-----	1,648
17		-----		-----		-----		-----	1	6,178		-----	1,506
21		-----		-----		-----		-----	1	3,942		-----	1,928
22	2	15,522	2	4,998	2	3,476	2	18,118		-----	1	14,424	2,424
25		-----		-----		-----		-----		-----		-----	1,674
27		-----		-----		-----		-----	1	12,672		-----	1,104
29		-----		-----		-----		-----	1	11,088		-----	1,870
30	1	12,672	1	4,754	1	4,176	1	9,178	1	7,374		-----	1,856
31	2	17,920	2	4,168	2	3,376	1	16,336		-----	1	1,584	2,418
August													
4	1	10,836	1	7,920	1	4,688	1	9,472		-----	1	9,404	1,752
5	1	14,256	1	8,942	1	6,658	1	17,424	1	8,972		-----	1,210
7		-----		-----		-----		-----	1	14,848		-----	1,972
8		-----		-----		-----		-----		-----	1	8,878	1,784
13		-----	1½	7,128	1½	4,728	1½	10,692	1	8,962		-----	1,152
14		-----		-----		-----		-----		-----	1	4,634	1,094
15		-----		-----		-----		-----	1	5,316		-----	886
Average		11,763		6,293		4,390		13,311		9,948		7,221	1,622

The highest count on the Pullman was obtained on August 5 when the concentration was five times that of the city. This was a day of excessive counts all through the train. Two days later the peak count on the electric train (eight times the average for the city) was obtained. On August 13 the peak count for the auto was recorded, and almost exactly the same count (seven times the city average) was found on the observation platform of the train.

On July 14, all the counts were low. Taking the count on the Pullman as unity, the following were the records obtained: coach, 1.1; locomotive, 1.37; observation platform, 2.34; and private automobile, 1.63.

The excessively high amounts of pollen encountered in this study are due, no doubt, to two factors: first, the speed of the vehicle actually "scooping up" the pollen from the air; and second, the vacuum action holding it there to settle.

These observations would seem to be sufficient to account for the experiences of hay fever sufferers who are compelled to travel.

SUMMARY

A study of the literature showed that the cause of hay fever varies widely in different sections of the country and that there is no uniformity in the atmospheric pollen concentration. It has been found, therefore, of increasing importance to make botanical surveys of the patients' environment, including the plant distribution and pollen production as well as the sensitivity of the patients. Many such studies have been made throughout the United States and Europe.

Blackley's studies, published in 1873 and 1880, are outstanding pieces of research on hay fever. To Blackley belongs the credit for the method of pollen determination now still in use and also the method of cutaneous testing for the sensitivity of patients. Scheppegrell in 1917 devised a formula for the determination of the pollen content of the air, in grains per cubic yard, and this has been used by many investigators, but, because ragweed pollen is the chief cause of hay fever, practically all the reports thus far published have stressed the ragweed pollen content of the air.

Since no reports were available regarding the wind-borne pollens of the Rocky Mountain region, I made botanical and air analysis studies in 1928, 1929, and 1930. Six test plots were chosen in which flora representative of the entire intermountain states was found; that is, alkali flat, desert, swamp, canyon, farming land, and high-grade residence property. From the results (Table I) it was evident that ragweed was neither the most abundant plant in the region nor the chief cause of hay fever as determined by cutaneous tests on hay fever patients. A total atmospheric pollen survey of

Utah as a typical intermountain state was undertaken, therefore, rather than merely a ragweed survey. Pollen counting stations were established in four Utah towns.

The results were graphed (Chart 1) and showed that the peaks of pollination fell in late July and early August when the grasses were beginning to wane and the weeds reached their maximum. Secondary peaks occurred in September when the sages pollinate.

The total pollen production was estimated for the plants found (Table II). The most important anemophilous plants of this region proved to be as follows: (1) *Graminaceae.* (2) *Chenopodiaceae*—Russian thistle, salt bush, summer cypress. (3) *Artemisia*—sages. (4) *Ambrosiaceae*—ragweed.

From cutaneous tests on 1,801 hay fever patients in this area, it was determined that 54.4 per cent of them reacted to the pollen of Russian thistle, 33.7 per cent to sagebrush and 31.4 per cent to common ragweed (Table III). The pollen abundance and the reactions of patients were compared, as below.

	POLLEN RATING	PER CENT REACTING TO 2 PER CENT POLLEN EXTRACT
Russian thistle	150	54.4
Sagebrush	130	33.7
Common ragweed	100	31.4
Timothy	40	26.4
Marsh elder	50	25.2
Blue grass	160	24.1
Box elder	35	21.1
Lambs quarter	20	19.1

A scratch reaction of 1:5,000 histamine was used as a positive control and no reactions from cutaneous scratch tests smaller than the histamine wheal were regarded as positive. Scrub oak was proved to have low toxicity. The box elder in spite of low abundance and pollen production accounted for 21.1 per cent of the patients. The fact that ragweed is not so important a factor in the hay fever of this region is due to its low distribution.

The sensitivity tests on patients are shown in Table IV which demonstrates the high percentage of patients sensitive to more than one pollen and the low percentage of those whose hay fever is due to a single pollen. There is a considerably higher percentage sensitive to weed pollen than to tree or grass pollen, while nearly a third are sensitive to more than one group.

In connection with this work an incidental study was carried on with regard to the effect of travel on hay fever symptoms. Pollen counts were made on vehicles of various types to determine the amount of pollen entrapped on petrolatum-coated slides. All counts were

equated to a twenty-four-hour basis for purposes of comparison. Using the counts obtained in Salt Lake City as unity the following ratios for pollen concentration were obtained:

Pullman coach	2.7
Chair car	3.8
Automobile	4.7
Electric train	6.2
Locomotive cab	7.2
Observation platform	8.0

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CASTOR BEAN DUST SENSITIZATION

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SENSITIZATION to organic dust in occupation has long been recognized as a cause of asthma and perennial hay fever. The literature cites many examples of poisoning due to ricin, a very irritating constituent of the castor bean but only a small number due to sensitization of the dust, causing allergic symptoms. Arnold[1] reported two cases of sensitiveness in which ingestion of the castor bean caused edema of the mouth, pharynx and in one case edema of the glottis. These symptoms occurred in five minutes. Both patients developed urticaria and were relieved by the subcutaneous injection of adrenalin. He also cited the case of a soldier who had eaten five castor beans for a cathartic, as a substitute for the oil. He died in coma due to nephritis caused by ricin poisoning. Snell's[2] patient had always handled castor beans with impunity; then developed violent sneezing, cough, puffy and reddened eyes, wheezing respiration and itching of the skin, on exposure to the castor bean. Robbins[3] also reported one, in a laboratory worker involved in research work with the castor bean. No skin tests were reported to have been done on any of these to determine the degree of sensitiveness.

Figley and Elrod[4] reported thirty cases of castor bean dust sensitivity and on investigation they found that some of these individuals were working in a linseed oil mill in Toledo, and others lived in the neighborhood of the mill. On further investigation it was found that the linseed oil mill not only manufactured linseed oil but also expressed castor oil from castor beans. After all the oil was removed from the beans, the residue was finely ground to form the castor bean pomace which was sacked and sold as an ingredient of fertilizer because of its high nitrogen content. The thirty patients were tested cutaneously by the scratch method with 1:100 dilution of the extract of castor bean dust, and every one gave a four-plus local reaction. Knowing that castor bean dust contains the very irritating substance, ricin, they made control skin tests on forty patients, many of whom had hay fever, asthma and perennial hay fever; all with negative results. Fifteen of their thirty patients gave a positive family history of hypersensitiveness. This compares favorably with the positive family history reported by Cook and Vander Veer—48.4 per cent, Spain and Cook—58.4 per cent, Coke—52 per cent, Rowe——56.4 per cent.

Vaughan[5] traced a case of hay fever and one of asthma in Suffolk, Virginia, due to castor bean meal which had come from Toledo and

which was used in the manufacture of fertilizer. These individuals worked in the fertilizer factory. He also reported a case of a boy, living in a small town in North Carolina, sensitive to castor bean dust and found that he handled fertilizer which was obtained from the factory in Suffolk.

Bernton[6] reported a case of a chemist who after three years' service with the U. S. Dept. of Agriculture, noticed that after exposure to castor bean dust, he suffered itching of the eyes, lachrymation, sweating of forehead, sneezing and coryza. The attacks were aggravated by attacks of asthma at night. There was constant exposure to dust, and the slightest inhalation of the powder produced attacks. Testing by the scratch method produced a marked local reaction with lymphangitis. A local reaction appeared with dilutions of the extract up to 1:250,000. No reactions were obtained with higher dilutions. He also tested eleven asthmatic individuals and six hay fever patients with the same extract, and the cutaneous tests were all negative. He did not attempt to desensitize the castor bean sensitive patient. The chemist was transferred to another laboratory and has been free from attacks of sneezing or asthma.

Follweiller and Haley[7] reported three cases developed in the research work with the castor bean and its products. In all three, there was marked improvement on withdrawal from the laboratory. No skin tests were done.

Barnard[8] reported seven patients who were sensitive to castor bean extracts and was able to remove the ricin present in these extracts without altering their activity.

Ratner and Gruehl[9] have shown that the toxic element, ricin, can be clearly differentiated from the allergic element. They reproduced the clinical picture in the guinea pig and assumed that the allergic reactions were related to bronchial asthma in the human being. They showed that sensitization could be induced by repeated inhalation of small amounts of dust, also anaphylaxis as demonstrated by the intravenous injection of the castor bean dust or pomace extract after an incubation period of seventeen days; that ricin death can be brought about by the inhalation of castor bean dust and immunity to ricin produced by repeated inhalations of small amounts of the dust after the pomace was partially dericinized. If due to anaphylaxis, Osborne, Mendel and Harris,[10] have shown that the animals die immediately with blanched and ballooned lungs whereas if due to ricin, death occurs within twelve hours after injection with congestion and diffuse hemorrhages of the lungs, hemorrhages of the abdominal viscera, involving Peyer's patches and the adrenals.

Case 1.—The case presented is of a white male, aged forty years, a longshoreman by occupation, engaged in unloading cargo from ships

coming from South America. He was admitted to the Allergy Clinic at the Long Island College Hospital in July, 1933. His chief complaints were itching of the eyes, sneezing and paroxysmal attacks of cough and difficult breathing. His family history did not reveal any allergic individuals. In his past personal history, he had had recurrent attacks of bronchitis at the age of four years, and pneumonia and malaria at the age of twenty years. A nasal operation and tonsillectomy were performed in March, 1933, without any relief from his symptoms. His employment as a longshoreman dates back to 1927. In 1928 he was told to eat a castor bean as a substitute for a cathartic and suffered with nausea, vomiting, abdominal cramps and diarrhea for forty-eight hours. In May, 1932, he noticed that exposure to castor bean dust, while unloading castor bean bags from a ship, was accompanied by sneezing, coryza, and itching of the eyes. The symptoms were aggravated by attacks of cough and wheezing respiration at night, and would last about twenty-four hours. Prior to May, 1932, he had been exposed to castor bean dust without any disturbance. Since then, every time he had been exposed he complained of sneezing, coryza, itching of the eyes and attacks of cough and wheezing respiration at night. At times the asthmatic attacks would occur at the time of exposure to the castor bean dust. Even moving about the sacks of castor bean at the far end of the pier, when he reported for work caused sneezing, coryza, and itching of the eyes. The severity of the symptoms depended on the amount of dust and the distance from the sacks.

Physical examination showed a well-developed Italian male patient. The conjunctivae were reddened. The nose showed polypoid middle turbinates and a toxic mucosa. There was no evidence of any tonsillar tissue. The chest wall showed slight excursion, and breathing was forcible. Percussion of the chest elicited a hyperresonant note with extension of the normal lung borders. On auscultation, expiration was prolonged with a relative shortened inspiration. Many sibilant and sonorous râles were audible. The apex beat was not palpable and the borders of the heart could not be percussed out because of the precordial emphysema. The cardiac sounds were barely audible. Examination of the abdomen revealed nothing of importance. Spleen and liver were not palpable. The reflexes were active and the finger nails showed a slight cyanosis.

X-ray examination of the chest showed that both lung fields were well aerated. The hilic densities and lung markings were moderate. The lung fields were negative for recent infiltrative changes. The blood Wassermann and Kahn reactions were negative. The white blood cell count numbered 9,000 with a differential of polymorphonuclear 70 per cent, small lymphocytes 21 per cent, large lymphocytes 6 per cent, eosinophiles 3 per cent.

Skin tests with the intracutaneous method were made against all common foods, pollens, and epidermal allergens. All these were negative except rye, which was labelled 1-plus, a 2-plus reaction to string beans, a 1-plus to celery, and a 2-plus reaction to banana. An intracutaneous test was then made with 1:100,000 dilution of castor bean dust extract. Immediately there developed a wheal with large irregular pseudopodia and its spread was very rapid. In order to check its further spread, it was necessary to inject epinephrin solution 1:1,000 at the site of the skin test, and in order to prevent absorption of the protein into the circulation, it was necessary to apply a tourniquet above the test and introduce another injection of adrenalin above the tourniquet. This was labelled a 4-plus reaction. A 4-plus reaction was also elicited with 1:1,000,000 dilution, a 3-plus with 1:10,000,000 dilution, and a 3-plus with 1:100,000,000 dilution. Twenty-four hours later an area of necrosis developed at the site of injection where the 1:100,000 dilution of the castor bean extract had been used. Repeated intake of castor oil by ingestion failed to produce any allergic or gastrointestinal symptoms. He has been free from attacks of itching of the eyes, sneezing, cough and wheezing respirations since he has avoided contact with castor bean dust by changing his occupation.

Case 2.—A male, aged thirty-four years, was employed as first mate on board ship. He gave a history of having had asthma for the past five years. His attacks occurred only when the ship's hatch was loaded with castor beans and the hatch was open. While at sea, the hatch was closed and the patient had no allergic symptoms. During the past six months, he had been assigned to shore duty. His work consisted of checking cargoes of castor beans. As a result, his attacks have occurred every day. With all the attacks of cough and wheezing respirations, he complained of nasal obstruction, itching of the eyes, reddened conjunctivae and an urticaria of the exposed surfaces; involving the face, neck and hands. The urticaria would appear immediately, if the castor bean dust which was present on the exposed surfaces was rubbed in with his hands. All intracutaneous tests were negative, except house dust, which showed a moderate reaction. Testing intracutaneously with a dilution of 1:1,000,000 of castor bean dust extract showed a 3-plus reaction.

SUMMARY AND CONCLUSIONS

1. Two individuals acquired a sensitiveness to the dust of the castor bean, characterized by sneezing, coryza, itching of the eyes, cough, wheezing respiration; and in addition in the second case, urticaria of face, neck and hands appeared.

2. Exposure to castor bean dust caused allergic symptoms, while avoiding it afforded complete relief.

3. Testing intracutaneously with castor bean dust extract in the following dilutions produced the following reactions: 1:100,000 4-plus; 1:1,000,000 4-plus; 1:10,000,000 3-plus; 1:100,000 3-plus; and the second individual gave a 3-plus reaction to a dilution of 1:1,000,000.

4. A review of the literature does not reveal a case of castor bean dust sensitivity which shows such marked positive skin tests in such high dilutions of the extract as reported in these cases.

5. Although other extracts of powerful allergens, such as horse dander, horse serum, rabbit epithelium, cottonseed, pollens and fish, are tested with in high dilution, castor bean extract requires still higher dilutions for testing in order to avoid constitutional reactions.

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Society Proceedings

THE ASSOCIATION FOR THE STUDY OF ALLERGY

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(CONCLUDED)

Observations on the Cytology of the Nasal Secretions in Allergy. FRENCH K. HANSEL, St. Louis, Mo. (For original article, see page 357.)

The So-called "Nonallergic" Vasomotor Rhinitis. HARRY L. HUBER, AND GEORGE F. HARSCH, Chicago, Ill. (Abstract.)

The authors maintain that about 70 per cent of cases of vasomotor rhinitis can be definitely identified as allergic in origin. The other 30 per cent present a difficult problem. A detailed study was made of such cases. This included a history, complete physical examination, examination by a competent rhinologist, and correction of significant nasal pathologic conditions, and the following laboratory examinations: blood count, blood chemistry, Arneth index, sedimentation rate, endocrine studies, nasal smears, skin tests, and x-ray examination of the sinuses.

From such studies the following data were assembled: 80 per cent are females, most of them between the ages of thirteen and fifty years. Ninety per cent have no known hereditary history of allergy. The majority have basal metabolic rates below –10 per cent, some with definite signs of myxedema. The majority have a low blood pressure and a slow pulse rate. The eosinophiles in the nasal smears are in about the same proportion as found in the typical allergic patient. Eosinophilia in the blood, although above normal, was distinctly lower than in the typical allergic patient. The blood chemistry and phosphorus are rarely abnormal. The sedimentation rate is usually a slow normal—distinctly more rapid than in the allergic patient. Skin tests are negative.

The frequent finding of definite evidence of endocrine dysfunction in many of this group of patients was based upon the following observations: (1) In the female, symptoms are frequently much worse at the menstrual time. (2) There is an unusual frequency of sterility without definite pathologic condition. (3) There is frequent association with the early months of gestation and often improvement in the last months. (4) There is frequent association with the menopause, normal or surgical.

There is a definite anatomical basis for the presence of the above mentioned symptoms. An abundant distribution of cavernous tissue in the nose is present. This tissue is usually considered an important secondary sex organ. It is engorged during menstruation and the early months of gestation, and explains vicarious menstruation. A definite neuron circuit between the pelvic organs and the nasal tissue has been established. Gynecologists frequently cauterize the genital spots in the nose in treating certain types of dysmenorrhea.

In this type of case the following measures have been used: Calcium lactate or gluconate up to 120 grains daily seemed to give relief occasionally. Urea nitrate in doses of 9 to 12 grains daily has given relief, sometimes striking, in about 30 per cent of the cases. Iodine was not beneficial. A general tonic program including diet and hygienic measures gave relief in many. Sphenopalatine ganglion injections gave temporary relief. Specifically, thyroid, or thyroid and parathyroid in small doses helped in a few cases. Antuitrin was not beneficial, but antuitrin "S" gave better results in some cases.

From these observations it is suggested that in the so-called "non-allergic" vasomotor rhinitis group, endocrine dysfunction may be considered as an etiologic possibility.

DISCUSSION

Dr. Francis M. Rackemann.—I am delighted to know that it is not all allergy, and I think it is quite right to emphasize that some of these cases may have a different fundamental background.

I would ask Dr. Huber if it is not true that most of his women patients were not fat rather than thin. If so, there is one little dodge that has helped my patients and that consists in making them lose weight by a reduction of their total dietary, and an increase of their outdoor exercise.

Dr. J. A. Rudolph.—There is confusion in my mind about some of the allergic conditions. I find this same confusion existing in considerable of the literature on this subject. We see patients who have had various diagnoses as Dr. Huber mentioned. We wonder whether all these differences cannot be classified.

We have been able to divide cases of nasal allergy into three distinct groups: the seasonal hay fever, the type we are all familiar with; the perennial hay fever, with or without some seasonal exacerbation, in which one can obtain a definite clinical history; and definite positive skin tests. Nasal smears show many eosinophiles and a few neutrophiles. With the above mentioned evidence one can make a diagnosis of perennial hay fever. These cases do well under proper instruction and therapy. The reports one sees of 75 and 85 per cent results in the treatment of nasal allergy are, in our opinion, in this group of cases.

On the other hand, we see another group on whom we get very poor results. This is the type of case we call vasomotor rhinitis or hyperesthetic rhinitis, and comes to us in considerable numbers because the rhinologist has tampered with their noses surgically and otherwise, with very little or no beneficial result. They are also very trying to us.

In this type of case we get a very indefinite clinical history, and our physical examination does not reveal any particular endocrine type. We see normal looking women and normal looking men, so far as the physical examination is concerned. We find no abnormalities about the skull or in the general examination, and skin tests usually reveal nothing, except perhaps some reactions of a plus-minus nature. We have tried elimination, trial and error diets, and we have tried therapy, nonspecific and specific. We have treated them with vaccines and have tried every conceivable type of glandular therapy, and calcium, but with very slight success.

Perhaps this type of case may have a different mechanism than the true perennial hay fever that we see.

Dr. H. P. Schenck, Philadelphia, Pa.—Dr. Hansel has done work which is so far superior to ours that we are ashamed to mention ours. Dr. Hansel takes a lot of pains in getting his smears, but we are much too impatient for that. Somebody suggested that if celophane handkerchiefs could be obtained and if the patients blow into them we could obtain more rapid specimens. We found that this worked fairly well. We could wash it quicker and transfer it to the centrifuge tube and stain the centrifuged material. There may be some objections to that, and I wanted to ask Dr. Hansel whether he saw any.

The other thing I thought might be worth mentioning is the fact that eosinophiles are being continually associated with nasal allergy. Frequently, because of the number of articles which mention this fact, inexperienced observers make smears from the nose, find no eosinophiles and conclude there is no allergy. We must remember that in seasonal allergy (seasonal hypersensitiveness) we may find eosinophiles during the attack; or if the discharge is too profuse, as Dr. Hansel has pointed out, it may be very difficult to find eosinophiles. However, between those attacks we may never find eosinophiles.

This applies to other forms of allergy besides the true seasonal types, because patients are not exposed to allergens all the time. There are periods of freedom. It is during those intervals we may not find eosinophiles!

Dr. J. Forman, Columbus, Ohio.—It seems to me that the majority of cases with vasomotor rhinitis, as has been said, are atopic in character. In the rest of them, it is best to look for something related to contact dermatitis. Some of them we see are sensitive to certain chemical substances, such as alcohol in any form. In these cases there is a nasal congestion in no matter what form they may attempt to ingest alcoholic beverages. I think we have to look upon these things as related to contact allergy.

When it comes to consideration of the secondary factors, it seems important not to emphasize the use of thyroid preparations too generally, because in my community it is quite the common practice to treat every one of these patients with large doses of thyroid without reference to allergy.

We do have a large number of girls who are decidedly undernourished, who have starved themselves, as any review of their diet will reveal. Those individuals have a natural defense mechanism set up in a low basal metabolic rate, which has nothing to do with disease of the thyroid gland. Those people had better be fed well, and the thy-

roid omitted. If thyroid therapy is to be used in vasomotor rhinitis to control secondary factors, it seems to me it ought to be done with the most careful selection.

DR. HARRY L. HUBER.—Dr. Rackemann, most of these patients are of normal weight.

DR. FORMAN.—I was careful to say that thyroid in small doses was given only in selected cases. I certainly agree with you that thyroid should be given only after careful study.

I want you to understand that not all of this group has a demonstrable endocrine involvement. It may be as we make more complete tests with more refined proteins, that some of these patients will be found to be allergic; however, there is much evidence to show involvement of the endocrines.

DR. HANSEL.—I am glad Dr. Schenck brought up that point about taking specimens on cellophane. Cellophane or wax paper is very good, particularly in children where one may have difficulty in swabbing the nose.

I forgot to mention that we have been using Jenner's stain in place of Wright's. We found it more reliable. The neutrophiles do not have the true granules, but it may be confusing to any one who has not examined a great many smears.

I might say that during very quiescent periods, without any apparent symptoms, often patients will show eosinophiles in the secretion, because it is perfectly possible for the nose to react in the form of a mild edema with the production of eosinophiles without any very definite symptoms.

I might say just a word about Dr. Huber's type of case. We have never been able to distinguish very clearly these cases from the others. Perhaps they represent a more hypersensitive type, particularly from the standpoint of the nose. If their nasal secretions show eosinophiles and they have these violent symptoms that other allergic patients have, I do not see how you can very definitely separate them from the allergic group.

DR. HUBER.—Dr. Rackemann asked the question about the obesity of the patient. Part of these were very obese, and others of them are practically normal weight.

Dr. Rudolph has somewhat the same findings as ours; about 20 per cent of them fall in the undiagnosed group.

Dr. Forman, I tried to be careful to say that in the specific treatment, thyroid in small doses in selected cases. I certainly agree with him that thyroid should be given only to selected cases.

We found that eosinophiles were present in an appreciable number, about 50 per cent of our patients, that is, of this whole group. I want you to understand that not all of this group falls in the endocrine class. That is only one part of it. As I said, it may be that as we make more complete tests, and with more refined proteins, we shall be able to diagnose many more of them than we are doing at present. However, as we look over them, we find there is not the hereditary history, and that there is a lower percentage of eosinophiles and some of the other things that do not quite fit into the typically allergic case.

Department of Reviews and Abstracts

Selected Abstracts

Anaphylactogenic Properties of Purified Milk Proteins, Raw, Evaporated, Superheated, Dried and Acidified Milk. Ratner, Bret, and Gruehl, H. L.: Pro. Soc. Exper. Biol. & Med. **31**: 559, 1934.

The immunochemistry of evaporated, superheated, freshly boiled, acidified and dried milks was studied to determine why individuals sensitive to raw or pasteurized milk can generally tolerate milks modified by heat.

Experiments upon guinea pigs showed that evaporated, freshly boiled and acidified evaporated milk, when fed, produced a markedly reduced sensitizing ability, whereas acidified raw, dried and superheated milks showed no difference from raw milk in this respect.

The presumption is that the loss in antigenic properties in heated milk is due to the coagulation of the whey proteins. Furthermore, heating facilitates complete digestion, thus diminishing the probability of the absorption of native proteins through the intestinal wall.

Prausnitz-Küstner Reaction With Sera of Ragweed Hay Fever Patients to Ragweed Carbohydrate Fraction. Caulfeild, A. H. W.: Proc. Soc. Exper. Biol. & Med. **31**: 573, 1934.

In a series of skin tests upon ragweed-sensitive hay fever patients, it was found an almost equal number of positive reactions was elicited by the protein and the carbohydrate fractions of ragweed pollen. Serum was then taken for the Prausnitz-Küstner reaction from (A) those giving the most marked reaction to the carbohydrate, (B) those giving negative reaction to the carbohydrate but strong positive reaction to the protein or whole pollen. With the serums of Group A, a local reaction to the carbohydrate solution could be elicited upon the sensitized area in the recipient, whereas no reaction was obtained with the serums of Group B.

The inference is drawn, not that carbohydrate functions as a true antigen, but that in a certain percentage of cases it has its own type of specific immunologic activity.

The Demonstration of Histogenic Antibodies in Allergic Skin Conditions. Perutz, A.: Wien. klin. Wchnschr. **46**: 1,023, 1933.

The three postulates with which it is usually considered that a skin disease should comply in order to fall under the classification of "al-

lergie'' are reviewed: (1) specificity of sensitivity; (2) the possibility of experimental sensitization; (3) passive transfer of sensitization.

It is with the third postulate, passive transfer, that there has been some uncertainty expressed, and it is with this postulate that the author chiefly concerns himself. He believes that in all passive transfer experiments a suitable control has been lacking. A series of unknown factors has been involved which could not be analyzed clinically or individually isolated. When the wheal content is passively transferred, it is not a matter of working with an individual body but with a fluid which contains not only specific substances but also a number of unspecific substances which do not fall in the category of antibodies.

In a series of experiments on iodoform dermatitis in 1928, the author showed that the reaction body fell in the group of thermolabile substances. Passive transfer could be obtained with the normal content of the wheal; transfer could not be obtained with wheal content that had been heated at 55° for one-half hour.

With a case of turpentine dermatitis that gave large wheals, the author inactivated half of the wheal content by warming at 55° for one-half hour; the other half wheal content was left active. Both fluids were injected next to each other on a normal control person, and tested with the allergen. Strongly positive reaction was obtained with the active fluid, negative reaction with the inactivated fluid.

He concludes that through heating, the thermolabile antibody is destroyed. The wheal content thus treated is then robbed of its antibody and so no longer reacts with its antigen. The control experiment, the author states, characterizes the reaction (passively transferred) as an antigen-antibody phenomenon.

Relation of Allergy to General Resistance in Streptococcic Infection. Clawson, B. J.: J. Infect. Dis. **53**: 157, 1933.

In this study the term allergy is used by the author to mean hypersensitiveness to a bacterial protein. The particular experiments reported have to do only with the streptococcic protein. The purpose of the study was to determine whether the state of allergy to streptococci bore any relation, either useful or harmful, to the process of general protective resistance against streptococci.

Young rabbits, free from any known infection, were used. The strain of streptococcus used in all the experiments was one recovered from the blood in a case of acute rheumatic fever. Record was made of the relative abilities of rabbits highly allergic, and rabbits highly resistant but with no evidence of allergy, to get rid of a known number of living streptococci. Normal animals were used as controls.

A series of rabbits was made allergic by subcutaneous injection of 10 c.c. of a mixture of equal amounts of a broth culture of streptococci and plain agar.

Another series of rabbits was made resistant (but not allergic) by intravenous injections of 5 c.c. of broth culture of streptococci at five weekly intervals.

Both series were subsequently tested for allergy by subcutaneous injection in ten different spots on the back of each animal of 0.1 c.c. of streptococci in saline solution. The number of organisms injected was so standardized that little or no reaction was elicited in normal animals.

The rate at which 50,000,000 living streptococci intravenously injected were removed from the blood (after 15 minutes) and from the liver (after two hours) was compared in those two series and in normal rabbits.

The experiments showed that organisms were not removed from the blood stream as rapidly in 15 minutes in the allergic animals as in the normal control animals. However, after two hours, a slightly greater number of streptococci per gram of liver were killed in allergic animals than in normal animals.

In resistant animals, organisms in the blood stream and in the liver were killed at much greater rate than was the case in either the allergic or the normal animals.

The conclusions are drawn that:

(1) Resistance to streptococcic infection can be developed artificially without the production of a measurable amount of allergy.

(2) The state of streptococcic allergy bears no necessary relation to general resistance.

(3) Allergy as related to general resistance to streptococci seems to bear no useful but rather at times a harmful rôle.

Negative Skin Reactions to Gland Extracts. Chapman, E. M.: Proc. Soc. Exper. Biol. & Med. **31**: 575, 1934.

Extracts from the lymph nodes of an untreated case of Hodgkin's disease and from a case of lymphosarcoma were used in skin tests upon eleven patients with Hodgkin's disease. In all but one patient, the reactions were negative. In the one patient, a wheal measuring 1.0 by 1.5 cm. at twenty-four hours and 1 to 2 mm. after forty-eight hours, was elicited by the extract in contrast to a saline and 0.3 per cent tricresol control measuring 0.5 cm.